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IRON METABOLISM IN INFANCY

I. Factors Influencing Iron Retention on Ordinary Diets[1]

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From the Harriet Lane Home of The Johns Hopkins Hospital and the Department of Pediatrics of The Johns Hopkins University School of Medicine

Submitted for publication April 13, 1939

In a paper published five years ago (1), a study of iron metabolism was reported on a group of six normal infants with the object of determining the quantitative relations between the actual amount of iron utilized by the normal infant for hemoglobin formation during the first six months of life, and the amount of iron available for hemoglobin formation derived from (1) the food, (2) the stores at birth, and (3) the "extra" hemoglobin with which the infant is provided at birth. It was concluded that under ordinary conditions the stores were utilized during this period of rapid growth to such an extent that they would become "exhausted" at about the end of the first half year of life. This, of course, did no more than confirm quantitatively what had been the accepted teaching for a long time.

In this work it was shown that during the first three months of life there was a greater tendency toward a negative iron balance than later, a fact also demonstrated by others who had studied iron balance. The total loss of iron, however, was relatively small, amounting to no more than about 4 mg. for the first two months of life. Recently Stearns and McKinley (2) have brought forth evidence to show that the iron loss in the first three months is much greater, their calculations bringing the total deficit to as much as 75 mg. This is a serious loss for an infant whose reserve of iron under the best of conditions rarely exceeds this amount. Several facts, however, give reason to doubt the correctness of their figures. (1) No other worker in the field has obtained negative balances approaching the magnitude of

[1] The expenses of this investigation were met by a grant from the Howland Memorial Fund.

theirs. (2) A loss of this amount would "exhaust" the stores at three months so that from this time on hemoglobin would be expected to fall, whereas all statistics from whatever source show a spontaneous rise in hemoglobin between the third and sixth months, and it is only after six months that the fall of hemoglobin that might be expected to follow "exhaustion" of stores, actually does occur. (3) They did not analyze the actual food that the baby took, and they collected the stools on cellu-cotton, in which the iron was determined and subtracted from the total; dubious procedures, as I know from experience.

Except in the work of Stearns and McKinley, no one has ever demonstrated a loss of iron from the body, under ordinary conditions in infancy, sufficient to be a major factor in the development of iron deficiency anemia. As was shown previously (1), however, the retention of food iron is nearly sufficient to cover the needs of growth after six months when the stores ordinarily become exhausted; and even when stores are exhausted as early as three months, a thing that might take place in premature babies or babies born of anemic mothers, it is impossible to explain, on a basis of iron deficiency, so severe an anemia as is often encountered without serious loss of iron from the body. The work reported in this paper is an attempt to study the influence of various factors on iron retention.

METHODS

The work was carried out on infants who, during the periods of study, were restrained on metabolism beds allowing the separate collection of stools and urine in porcelain or glass containers. Some of the infants used for long continued studies were thus restrained five days out of each week for six months without evidence of any deleterious effect on growth or muscular development. Most of the periods were four or five days, and were not specially marked, since the bowel movements were fairly regular.

Aliquots of each day's feeding were mixed together, and at the end of each period a sample was analyzed for iron. In the case of orange juice and farina, a few samples were analyzed and the average used to calculate iron from this source.[2]

[2] I do not believe it is advisable to use prepared tables for calculating iron intake in estimations of balance. The figures may be adequate for the averaging of several observations, but my own experience with the variability of the iron content of different samples of dried milk from the same source, makes me wary of tables. If I had assumed anything like an average iron content of dried milk in this work the results would have been altogether false.

The methods of stool and urine collection, preparation for analysis, and analysis have been previously described in detail (1). When it was found that the amount of iron in the urine was insignificant, being not much more than 5 per cent of the total output, determinations of iron in the urine were no longer carried out.[3] The total stools for the period were dried, powdered, weighed and a sample taken for analysis. Ashing was carried out in platinum dishes in an electric oven, the ash dissolved in HCl, and the analysis made colorimetrically by the thiocyanate-amyl alcohol method as previously described. The method used was modified from that of Kennedy and is not very different from that described by Stugart (3).

There are countless methods for iron determination, many of them differing from one another only in detail. Of these, the method of choice should be one requiring as little manipulation as possible. It is for this reason that the method used here included dry ashing and determination of the iron in the ash solution, without further attempt at purification. Much has been written of the effect of phosphates in preventing the full development of the thiocyanate color, and many investigators have attempted to remove the phosphates before the colorimetric analysis. In my experience this is unnecessary when amyl alcohol is used. The only precautions to be observed are the heating and subsequent cooling, immediately before colorimetry, of the solution to be analyzed, and the use of a standard freshly diluted from the concentrated stock solution of $FeCl_3$. The precipitation of phosphates in the attempt at purification is likely to lead to loss of iron with the phosphate precipitate; while the precipitation of iron may be incomplete owing to the formation of a colloidal iron solution. Moreover, the reagents used always contain iron. When the amounts of iron are as small as they are in biological material, the sources of error are considerable. This whole question is well treated in Stugart's article.

RESULTS

In the previous paper (1) it was shown that iron balance had little bearing on changes in hemoglobin during the first six months of life. The hemoglobin fell and rose again, depositing iron in the stores and utilizing iron from the stores, quite independently of the iron that was being ingested in the food. The changes that were going on in the body were, however, mirrored by small changes appearing, as it

[3] In the previous paper on iron metabolism (1), urine was included with the stools for iron determinations, because in that paper I was interested in balances. But even in that paper the conclusions would not have been altered by disregarding the iron in the urine; and the determinations were made more out of deference to tradition than because they were of any importance. In this part of the work no attention is paid to the urine.

TABLE Ia

*Infants under 3 Months of Age**

CASE NUMBER	AGE	HEMOGLOBIN	BODY WEIGHT	DURATION OF PERIOD	IRON INTAKE PER KG. PER DAY	IRON RETENTION PER KG. PER DAY	IRON EXCRETION PER KG. PER DAY	
		gm. per 100 cc.	*kg.*	*days*	*mg.*	*mg.*	*mg.*	
1	3 w.	20.6 falling	3.7	4	.13	−.06	.14	Normal infant
	4 w.	17.3 "	3.9	4	.16	+.04	.06	
	5 w.	16.0 "	4.1	6	.13	−.06	.14	
	6 w.	14.3 "	4.4	5	.14	−.014	.10	
	7 w.		4.5	5	.14	−.055	.14	
	8 w.	11.9 "	4.6	5	.15		.085	
	9 w.		4.8	5	.15	−.01	.10	
	10 w.	10.7	4.9	5	.14	+.02	.06	
	11 w.	10.4	5.2	5	.19	+.05	.06	
	12 w.			5	.20	+.04	.08	
	13 w.		5.5	5	.20	+.01	.11	
2	3 m.	10.7	3.3	4	.22	−.17	.30	Normal infant
3	4 w.		4.5	5	.11	−.11	.18	Normal infant
	5 w.		4.7	6	.11	−.02	.09	
	7 w.		5.0	10	.25	+.03	.12	
	8 w.		5.6	8	.21	−.03	.15	
	10 w.		5.9	5	.35	+.01	.20	
	11 w.		6.2	6	.20	+.06	.06	
	13 w.		6.5	7	.16	+.04	.055	
4	3 m.		6.1	5	.28	.03	.14	
5	2 w.	16.2	3.5	4	.08	0	.05	Normal infant
	3 w.		3.7	4	.09	0	.055	
	5 w.	12.0	4.0	4	.13	.03	.05	
	6 w.		4.4	5	.24	.01	.13	
	8 w.	10.5	4.7	5	.12	−.03	.10	
	9 w.		4.9	5	.13	−.04	.12	
	10 w.		5.0	5	.13	−.04	.12	
	11 w.		5.1	5	.16	+.06	.09	
	12 w.		5.3	5	.14	−.01	.095	
6	3 w.		3.6	4	.16	+.03	.07	Normal infant
	4 w.		3.8	4	.15	+.05	.04	
	5 w.		4.1	5	.14	+.025	.06	
	6 w.		4.3	4	.12	−.12	.19	
	7 w.		4.6	5	.12	−.08	.15	
	8 w.		4.8	5	.12	−.07	.14	
	9 w.		5.0	5	.18	−.045	.15	
	10 w.		5.3	5	.14	−.08	.16	
	11 w.		5.5	5	.14	−.03	.11	
	12 w.		5.8	5	.13	+.03	.05	

TABLE Ia—*Continued*

CASE NUMBER	AGE	HEMOGLOBIN	BODY WEIGHT	DURATION OF PERIOD	IRON INTAKE PER KG. PER DAY	IRON RETENTION PER KG. PER DAY	IRON EXCRETION PER KG. PER DAY	
		gm. per 100 cc.	*kg.*	*days*	*mg.*	*mg.*	*mg.*	
7	6 w.	15.0	2.4	4	.37	+.07	.15	Premature
	7 w.		2.8	4	.42	+.10	.15	Immediately after 5 day iron period
8	3 m.+	5.6 falling	2.0	4	.25	+.07	.08	Sepsis with hemolytic anemia
	3 m.+	4.3	2.4	3	.29	+.095	.08	Shortly after 5 day iron period
9	3 m.+	9.9	2.8	4	.24	+.05	.09	Premature twin
		12.2 rising	3.2	3	.26	+.095	.06	Shortly after 5 day iron period
10	2½ m.	10.9	3.4	5	.15	−.02	.11	
	3 m.	10.2	3.9	5	.28	−.09	.26	2 weeks after 5 day iron period
11	4 m.	14.3 falling	6.5	5	.15	−.06	.15	
		14.7	6.9	3	.19	+.005	.11	Shortly after 5 day iron period
12	3 m.	12.3	5.4	5	.18	−.02	.13	
13	3 w.	6	3.2	3	.30	−.02	.20	Hemolytic anemia of newborn
		6.5	3.3	3	.30	+.025	.155	
		7.0 rising	3.4	3	.30	+.12	.06	
14	6 d.	15.5 falling	2.3	5	.19	−.105	.22	Premature. No viosterol
	3 w.	9.7 "	2.5	3	.24	0	.145	
	1 m.	8.6	2.8	3	.27	+.003	.16	Immediately after 5 day iron period
	1½ m.	10.5 rising	3.6	5	.24	+.03	.115	
	2 m.	10.3	3.8	5	.21	0	.125	
	2½ m.	10.5	4.5	4	.27	+.08	.08	
	3 m.	10.5	4.8	5	.26	+.075	.08	
15	6 w.	12.6 falling	3.4	5	.32	+.065	.13	
	2 m.	13.0 rising	3.8	4	.24	0	.145	Immediately after 5 day iron period
16	2 m.	10.5	4.2	5	.21	+.008	.12	

TABLE Ia—*Concluded*

CASE NUMBER	AGE	HEMOGLOBIN	BODY WEIGHT	DURATION OF PERIOD	IRON INTAKE PER KG. PER DAY	IRON RETENTION PER KG. PER DAY	IRON EXCRETION PER KG. PER DAY	
		gm. per 100 cc.	*kg.*	*days*	*mg.*	*mg.*	*mg.*	
17	3 w.	21.4 falling	3.7	5	.27	+.05	.11	Normal infant
	1 m.		4.1	4	.32	+.08	.11	
	1½ m.		4.3	4	.30	+.06	.12	
	2 m.	13.2	4.8	4	.29	+.115	.06	2 weeks after 5 day iron period
	2½ m.	12.9	5.1	5	.16	−.04	.135	
	3 m.	13.3	5.6	4	.22	0	.13	
18	1 w.	16.5 falling	3.6	5	.33	+.18	.02	
	3 w.	12.2 falling	3.8	5	.30	+.081	.10	
	4 w.		4.0	5	.31	+.13	.055	
	6 w.	9.9	4.1	5				
	7 w.	9.7	4.3	5	.37	+.18	.04	
	8 w.		4.5	5	.36	+.07	.14	
	2 m.	9.7	4.6	5	.35	+.01	.20	
	2 m.+		4.7	5	.36	+.11	.105	
	2½ m.	12.0	4.9	5	.35	+.07	.14	
	2½ m.+		4.9	5	.29	−.06	.23	
19	4 w.		2.9	3	.30	+.06	.12	$CaCO_3$ added to milk in all periods
			3.0	3	.29	+.04	.135	
	5 w.		3.1	3	.31	+.01	.17	
			3.2	5	.27	−.03	.19	

As explained in the text, this table is not a balance sheet such as is usual in articles on iron metabolism; it merely presents the data which are necessary to the purpose of the article. Anyone interested may calculate the observed values from the given data since the body weight and the duration of the period are included. The derivation of the formula for calculating *excretion*, and the significance of this term, are explained in the text. *Retention* is calculated by subtracting the *output* in the stools from the *intake*. The *output* is not included in this table but may be calculated by subtracting *retention* from *intake*.

TABLE Ib

Infants 3–6 Months of Age

CASE NUMBER	AGE	HEMOGLOBIN	BODY WEIGHT	DURATION OF PERIOD	IRON INTAKE PER KG. PER DAY	IRON RETENTION PER KG. PER DAY	IRON EXCRETION PER KG. PER DAY	
	mos.	*gm. per 100 cc.*	*kg.*	*days*	*mg.*	*mg.*	*mg.*	
20	5		6.5	4	.185	+.004	.105	Normal infant
	5½		6.7	4	.21	−.09	.22	
	6		6.7	4	.18	+.018	.09	
5	3+		5.5	5	.14	+.005	.08	Normal infant
	3½		5.7	5	.13	−.012	.09	
	4		5.9	5	.14	−.012	.095	
3	3		6.5	7	.16	+.04	.055	Normal infant
	3+		6.6	4	.18	+.042	.065	
	3½		6.7	8	.048	−.015	.045	
	4		6.8	9	.079	+.01	.04	
2	3+	9.8	3.8	4	.20	.036	.085	Normal infant
	3½		3.9	4	.21	.032	.095	
	4−	9.9	4.0	4	.20	.05	.07	
	4	10.4	4.3	6	.20	.042	.08	
	4+	11.3	4.5	5	.16	.016	.08	
	4+		4.8	5	.18	.046	.06	
	4½	11.6	5.1	5	.18	.047	.06	
	5−		5.2	5	.20	.042	.08	
	5	11.7	5.3	5	.17	.049	.055	
	5+		5.3	5	.11	.019	.045	Breast milk
	5½		5.4	5	.11	−.004	.07	" "
	6−		5.5	5	.20	.029	.09	
	6		5.6	5	.14	.068	.02	
1	3		5.6	5	.17	.007	.095	Normal infant
	3+		5.8	5	.13	−.048	.125	Infection
	3½		5.9	5	.16	−.014	.11	"
	4−		6.0	5	.13	−.03	.11	"
	4	12.9	6.3	5	.15	+.017	.08	
	4+		6.5	5	.17	+.012	.09	
	4½		6.7	5	.15	.024	.065	
	5−		6.8	5	.20	.068	.05	
	5	12.0	6.9	5	.18	.024	.085	
	5+		7.1	5	.16	.017	.08	
	5½	13.4	7.2	5	.15	.019	.07	
	6−	12.0	7.4	5	.14	.019	.065	

TABLE Ib—*Concluded*

CASE NUMBER	AGE	HEMOGLOBIN	BODY WEIGHT	DURATION OF PERIOD	IRON INTAKE PER KG. PER DAY	IRON RETENTION PER KG. PER DAY	IRON EXCRETION PER KG. PER DAY	
	mos.	*gm. per 100 cc.*	*kg.*	*days*	*mg.*	*mg.*	*mg.*	
12	4−	13.2	6.0	3	.22	.011	.12	Immediately after 5 day iron period
	4	13.1	6.5	5	.16	−.031	.125	2 weeks after 5 day iron period
	5−		7.3	5	.25	+.038	.11	2 weeks after 5 day iron period
21	5−	11.6	4.8	4	.17	.027	.075	Scurvy?
	5+	12.6	4.9	4	.17	.055	.050	Infection
	5½	10.6	5.0	4	.17	.086	.015	
14	3+		4.8	5	.26	.075	.08	No viosterol
	3½−	11.0	5.1	4	.25	.072	.08	
	3½+		5.3	4	.22	.026	.105	
	4−		5.5	4	.18	.073	.035	
	4		5.7	4	.20	.060	.06	
	4+		5.9	4	.19	.024	.09	
	4½	11.5	6.1	4	.23	.048	.09	Viosterol begun
	5−		6.3	4	.21	.078	.05	
	5		6.5	4	.18	.052	.055	
	5½		6.7	5	.27	.102	.06	
17	3½		6.1	4	.23	.033	.105	
	3½+	15.3	6.3	4	.22	0	.13	
	4−		6.6	4	.25	.10	.05	Viosterol begun
	4	14.1	6.8	4	.25	.096	.055	
	4½	13.6	6.9	5	.26	.12	.035	
	5−	15.3	7.3	4	.25	.089	.06	Immediately after 5 day iron period
	5+	15.3	7.6	4	.25	.066	.085	
10	3+	10.2	3.9	4	.28	−.09	.26	After 5 day iron period
	4−		4.3	5	.25	+.028	.12	No viosterol
	4		4.7	5	.19	0	.115	
	4+	10.7	4.8	5	.16	−.006	.10	
	4½+		4.9	5	.19	+.041	.075	
	5	12.2	5.0	4	.18	.018	.09	
	5+		5.2	4	.23	.049	.09	
	5½	11.1	5.4	4	.22	.005	.125	
	6−		5.6	5	.24	.054	.09	Viosterol begun
	6	10.6	5.9	4	.22	.07	.06	

TABLE Ic

Infants over 6 Months of Age

CASE NUMBER	AGE	HEMOGLOBIN	BODY WEIGHT	DURATION OF PERIOD	IRON INTAKE PER KG. PER DAY	IRON RETENTION PER KG. PER DAY	IRON EXCRETION PER KG. PER DAY	
	mos.	*gm. per 100 cc.*	*kg.*	*days*	*mg.*	*mg.*	*mg.*	
10	6	10.6	5.9	4	.23	.071	.065	
	6+	10.7	6.2	4	.24	.068	.075	
	6½		6.1	5	.25	.089	.06	
	7−	12.4	6.4	4	.23	.078	.083	Immediately after 5 day iron period
	7	12.2	6.5	4	.23	.085	.05	
2	6+	11.5	5.6	5	.17	.057	.045	
	6½		5.7	5	.19	.035	.08	
	7−		5.7	5	.16	0	.095	
	7	11.2	5.8	5	.17	.024	.08	
12	8½		10.9	4	.19	−.055	.19	Immediately after 5 day iron period
22	6	11.8	5.2	5	.26	−.03	.185	
	6½	12.0	5.9	4	.24	0	.145	2 weeks after 5 day iron period
21	6	7.9	5.1	4	.16	.008	.105	Scurvy infection
	6½	8.9	5.1	4	.18	.043	.065	
	6½+	7.9	5.3	4	.12	.04	.03	
	7−	8.3	5.4	4	.13	.002	.075	
	7	7.1	5.5	4	.12	.04	.03	
	7½	6.9	5.8	4	.17	.086	.015	
	8	6.5	6.0	5	.13	.03	.05	
23		4.5	8.0	3	.19	.035	.08	Nutritional anemia
		4.3	8.0	6	.28	.14	.03	
24	19	5.5	8.2	4	.18	.001	.11	Nutritional anemia
25	17	8.5	7.9	4	.12	.05	.02	Post infection
26	18	6.9 falling	7.9	4	.105	.03	.03	Infection
		6.0	8.0	4	.14	0	.085	
27	10	7.9	8.0	4	.19	−.14	.25	

TABLE Ic—*Concluded*

CASE NUMBER	AGE	HEMOGLOBIN	BODY WEIGHT	DURATION OF PERIOD	IRON INTAKE PER KG. PER DAY	IRON RETENTION PER KG. PER DAY	IRON EXCRETION PER KG. PER DAY	
	mos.	*gm. per 100 cc.*	*kg.*	*days*	*mg.*	*mg.*	*mg.*	
28	14	3.5	9.1	3	.20	.11	.01	Nutritional anemia
		5.0 rising	9.8	5	.20	.10	.02	2 weeks after 5 day iron period
		5.2	10.5	5	.18	.015	.09	2 weeks after 5 day iron period
29	8	7.1	5.2	4	.25	.08	.07	Post-infection
30	8½	5.9	6.8	4	.25	.10	.05	Post-infection
	9	5.6	7.3	4	.16	0	.095	
31	10	7.3	8.8	5	.11	.028	.04	Post-infection
32	11	6.9	6.6	5	.22	.064	.07	
33	7½	13.7	6.4	5	.16	−.038	.135	Diarrhea
		12.6	7.2	3	.20	−.08	.13	
34	9	9.6	5.8	5	.15	.033	.055	Infection
	10	10.0	6.5	5	.20	.026	.095	2 weeks after 5 day iron period
	10½	8.9	7.0	4	.21	.079	.05	1 week after 5 day iron period
35	8	8.5	5.4	5	.20	−.045	.165	Osteogenesis imperfecta
	8½	8.5	5.5	4	.16	−.04	.135	1 week after 5 day iron period
	9	8.5	5.6	3	.16	−.04	.135	

were on the surface, as for instance in the frequent occurrence of a negative balance during the first three months of life, when the hemoglobin was falling. In the present work an attempt has been made to study these rather minute changes, in the hope that some light may be thrown on the more important internal iron metabolism, especially in relation to anemia. The Tables have, therefore, been prepared to present the data essential to this purpose, and they are in no sense balance sheets. Intake and retention of iron are recorded in mg. per

day per kg. body weight, instead of in absolute amounts for the period of observation.

1. Relation of retention to intake of iron. (Charts 1 and 2)

In spite of the scattering of figures which is to be expected in all work on iron balance, there appears to be a linear relation between

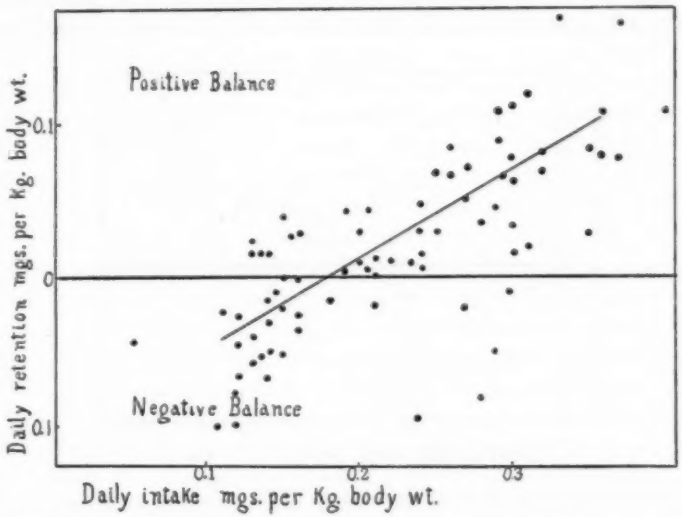

CHART 1. RELATION OF IRON RETENTION TO IRON INTAKE IN INFANTS UNDER 3 MONTHS OF AGE

The sloping straight line represents the average retention. In drawing this line the chart was divided into vertical sections which overlapped so that the right half of one section formed the left half of the next. The figures for retention lying within each section were then averaged, the point representing the average being placed on the vertical line passing through the middle of the section.

intake and retention, expressed by the straight line drawn through the points representing averages. It is evident from Chart 1 that about 0.17 mg. of iron per kg. body weight per day must be ingested by the infant of less than three months to bring the intake and output into equilibrium, and that if less than this is given the tendency is toward a negative balance. This touches one of the controversies of

the present time. According to Lintzel (4), confirmed by Farrar and Goldhamer (5), and by Reimann, Fritsch and Schick (6), there is no lower limit to the possibility of equilibrium in a normal adult. One of Lintzel's subjects was in equilibrium on less than 1 mg. of iron per day. Reznikoff (7), on the other hand, found a lower limit to positive balances at an intake of about 10 mg. per day, corresponding to about 0.15 mg. per kg. per day. Likewise Ohlsen and Daum (8) and Leverton and Roberts (9) found a point below which the balance tended to

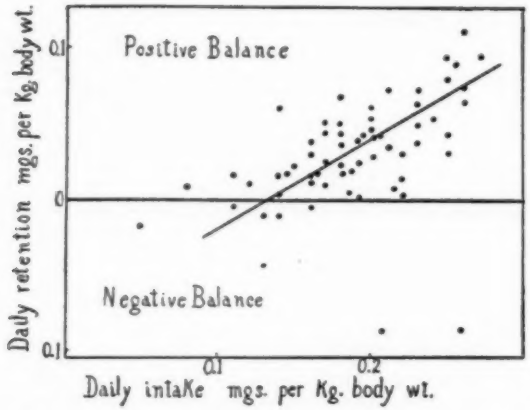

CHART 2. RELATION OF IRON RETENTION TO IRON INTAKE IN INFANTS FROM 3–6 MONTHS OF AGE

As may be seen, the slope of the curve representing average retention is the same as in the previous chart but the line lies at a higher level. This has been interpreted to mean that average absorption is the same in the two age groups, while excretion differs.

be negative, at an intake of about 0.15 mg. per kg. per day or slightly higher.

It is possible to reconcile these divergent opinions if one takes into account the character of the diet used; for to obtain the low intake figures the iron was largely derived from dairy products and cereals, whereas the second group was studied while on a general diet. Recent work at the University of Wisconsin (10, 11) and elsewhere (12) has suggested that the iron of foods varies in its "availability," that of

vegetables and fruits being relatively "unavailable," while that of cereals is much more "available." It is quite possible then that when a general diet is given, a greater intake of iron is necessary to obtain equilibrium, than when the diet is restricted to cereal and milk.

This may explain discrepancies in the amount of iron which different authors consider necessary for equilibrium but it does not settle the question of a lower limit. Lintzel is the only one who has found equilibrium on intakes below 5 mg. per day in the adult, the other workers not having gone below this figure. Rose, Valteich, Robb and Bloomfield (13), in a child of 2½ years, found negative balances at an intake of 0.33 mg. per kg. per day, when about 75 per cent of the iron was derived from peas and prunes. Daniels and Wright (14) and Ascham (15) studying preschool children found slightly positive balances at about 0.6 mg. per kg. of body weight, most of the children being on a general diet, but some on milk and cereal.

Chart 2, representing infants from three to six months, confirms Chart 1 in indicating that there is a level of intake below which the balance tends to become negative. This tendency to a negative balance on low intake is found also in the articles of Wallgren (16) and of Snelling (17), but is not found in that of Stearns and Stinger (18).

In spite of the rather marked scattering there is a trend in the figures which is indicated by the diagonal line which represents averages (see note, Chart 1). Though there may be objections to drawing such a line when the scattering is so great, it is used only to indicate a trend in the relationship of retention to intake of iron, and to introduce a method of graphic representation which simplifies the presentation of the observations. Before describing the latter it is necessary to say a few words concerning the problem of absorption and excretion.

When iron is ingested not all of it is absorbed, for a certain portion passes into the stools. In addition to this, some iron is excreted into the bowel, so that the iron in the stools has two origins (1) the unabsorbed iron and (2) the "excreted" iron. The term *iron excretion* in this paper refers only to the second of these categories; the term *iron output* refers to the total iron appearing in the stools. *Iron retention* refers to the difference between intake and output, while *iron absorption* refers to the iron actually taken up by the body from the gastrointestinal tract.

These statements may be expressed in the form of equations:

Retention = intake minus output

Excretion = absorption minus retention

The presence of the excretion which is going on all the time regardless of intake, is what is responsible for the negative balance on low intakes, for before the balance can become positive the excretion must be overcome by the absorption. If there were no excretion there would be no negative balance and the average curves of Charts 1 and 2 would touch the point of zero retention when no iron was ingested. Absorption would then equal retention, and it is easy to see that the slope of the curve would represent the *average* percentage of ingested iron that was absorbed. The fact that the average line tends to be straight is of importance, for to obtain a straight line either the *average* excretion must be more or less constant, in which case the slope of the curve will represent a constant percentage absorption, or else, if percentage absorption rises with increased intake, the *average* excretion must rise at the same rate. From all we know of iron metabolism the latter possibility is most unlikely; in the next paper it will be shown that "excretion" of iron is uninfluenced even by very large increases in iron intake. I have, therefore, accepted the former possibility, with the conclusion that *average* absorption represents a constant proportion of intake.

A certain confirmation of this point of view is found in Charts 1 and 2; for the slope of the line representing averages is the same in the two charts, the difference between them residing in the fact that the probable excretion is greater in the cases represented in Chart 1 than in those of Chart 2. But Chart 1 represents babies from birth to three months when hemoglobin is falling, and one would expect a greater "excretion" of iron as a result of breakdown of hemoglobin.

If one may then assume that average absorption bears a constant relation to intake, one has a means of determining average excretion from the equation given above:

Excretion = absorption minus retention

Since a measurement of the slope of the curve in Charts 1 & 2 shows that absorption is 60 per cent of intake, the equation becomes

Excretion = (intake $\times$ 0.6) minus retention

This of course represents average excretion, but if we take one more step we obtain a simple method of representing the results, which seems to have advantages over the usual method. This step is to calculate the individual excretions from the above equation. At first sight this seems unjustifiable since in the individual case it is not known to what extent absorption deviates from the average, and I may appear to be claiming that all the deviations from the average are dependent upon differences in excretion. I make no such claim, and merely use this method of expressing the relationship between retention and intake in terms of excretion, which in this case does no more than give a convenient method of denoting how far the points in Charts 1 and 2 deviate from the average. For instance, a calculated daily excretion of 0.1 mg. per kg. means that the corresponding point in Chart 1 will be on the line denoting the average.

In other words, deviations from the average are calculated by a method that expresses them in terms of excretion. Actually the deviations may depend as much on differences in absorption but it is easy to keep this in mind in attempting to interpret the results in the individual case. Whether we conclude that in a given case excretion is increased or absorption diminished, or *vice versa*, will depend on our knowledge of the factors ordinarily influencing these processes. In the period of falling hemoglobin previously cited, it is altogether probable that the excretion is increased; in the case of HCl medication, which is said to increase retention, it is just as probable that absorption is increased though the figures obtained by this method of calculation would be expressed as decreased excretion.

In Chart 3 the calculated excretion has been charted against age. The greater excretion of the first three months of age is well shown. After about the fourth or fifth month the figures appear to be fairly constant but there are too few observations in the older age groups to be certain of constancy. The figures already quoted for children over two years of age make it seem likely that excretion is greater or absorption less in this age-period, but the diets are not comparable and the importance of availability of iron in relation to diet has already been indicated. The figures given by Schlutz, Morse and Oldham (19) for periods when spinach and apricots furnished a considerable pro-

portion of the iron intake, and those given by Stearns and Stinger (18) for spinach and egg yolk indicate what is probably a considerable decrease in absorption when these foodstuffs are present.

2. Retention of iron and rise or fall in hemoglobin

The most obvious explanation of the higher figures for iron excretion in the first three months of life, is that they are the result of the large amount of iron set free from hemoglobin during this period. It is impossible, however, to find any correlation between the amount

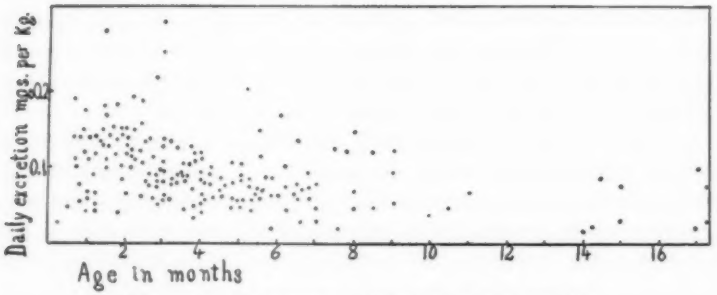

CHART 3. RELATION OF IRON EXCRETION TO AGE

The method of calculating excretion is given in the text. It must be understood that this method of formulating the results of the study of iron balance is used for its convenience. In the individual case it is impossible to be certain that the particular figure for retention is conditioned by differences in absorption or excretion. A low retention may be due to diminished absorption or increased excretion. This should be borne in mind in using this method (see text).

of excretion and the presence of a rising or falling hemoglobin. Three cases of hemolytic anemia were studied. In two there was no increased excretion of iron; in the third, a case of hemolytic anemia of the newborn, a fairly high excretion gave place to one within normal limits as the hemoglobin started to rise.

3. Prematurity

The observations in this work indicate that prematurity has no influence on iron retention. The cases that were studied, however, cannot be considered to settle the question because of the relatively

good birth weight (over 2.2 kg.). Snelling's (17) observations are more to the point but his figures were obtained while the babies were on breast milk and there are no comparable figures in my work. Wallgren (16), however, studied normal babies on breast milk and a calculation of his and Snelling's data give identical excretion figures. It is, of course, well known that Lichtenstein (20) claimed to have shown that premature babies had a tendency to a negative balance. He, however, used the old Neumann method for iron analysis which

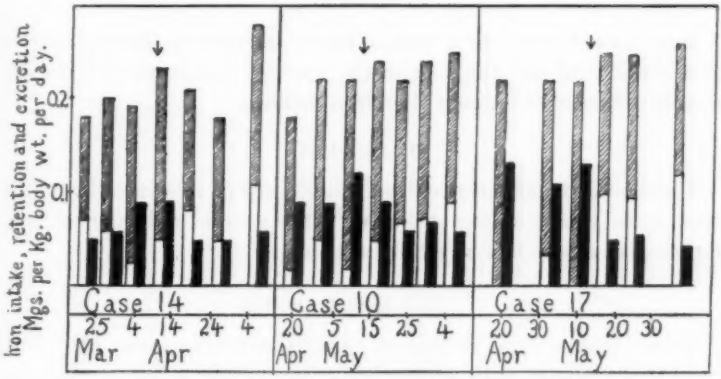

CHART 4. EFFECT OF VITAMIN D ON IRON EXCRETION

The left hand column of each pair represents intake, the unshaded portion at the bottom denoting retention, and the shaded portion representing the output in stools. The output in the urine is negligible. The black column represents iron excretion. The arrows point to the day on which viosterol was begun. When viosterol was given early signs of rickets were demonstrable by x-ray.

has been shown to be grossly inaccurate for small amounts of iron (Lintzel and personal observation).

4. Vitamin D deficiency

The effect of vitamin D deficiency was studied in three infants. Vitamin D was omitted from their feedings until the first signs of rickets appeared in the roentgenograms. Viosterol was then given. The results are to be seen in Chart 4. It is evident that about the time at which the roentgenograms showed signs of beginning rickets,

the iron excretion was above the average, and that viosterol caused an immediate fall in the excretion figures. The effect has been stated in terms of excretion, but it seems more likely that the factor that conditions the variation in retention, is absorption.

Although these changes are demonstrable, their magnitude is such as to have little practical bearing on the problem of total iron deficiency in the human. The problem of iron balance in clinical rickets still remains to be settled.

5. Vitamin C

Two cases of scurvy were studied before and after the administration of ascorbic acid and the cure of the scurvy. There was no demonstrable influence of vitamin C on iron balance.

6. Infection

This had little influence on iron balance except that infection might reduce the intake of iron along with the general reduction of food intake and so lead to a negative balance.

7. Nutritional anemia

In one case, anemia developed under observation (No. 21). This child was admitted with a diagnosis of scurvy, for which he had been treated before the study of iron metabolism was started. As may be seen, the hemoglobin fell relatively rapidly at first, then more slowly reaching 6.5 gm. per 100 cc. two months later. The cause of this anemia was not determined, an occasional mild infection hardly accounting for so striking a reduction in hemoglobin. During the period of falling hemoglobin, the iron balance was always positive so that it is evident that the iron which was set free was stored in the body. At the end of the first rapid fall in hemoglobin there was the rise in excretion which one might expect. When the patient was treated with medicinal iron, only a small portion of that which was retained, went to form hemoglobin; the rest being stored. (See next paper for figures.)

During the first six months of life the stores of iron have presumably been reduced to a low level owing to the demands of growth. Fontès and Thivolle showed in dogs whose iron stores had been re-

duced by successive bleedings, that iron retention progressively increased. It is possible, therefore, that the condition of the stores is a factor in the retention of iron. That this is not generally so is shown by the few figures for excretion in normal babies after six months of age.

On the other hand, in the cases of nutritional anemia studied, and in Case 21, during quiescent periods the excretion of iron tended to be below the average, i.e., the body tended to retain the iron. This is

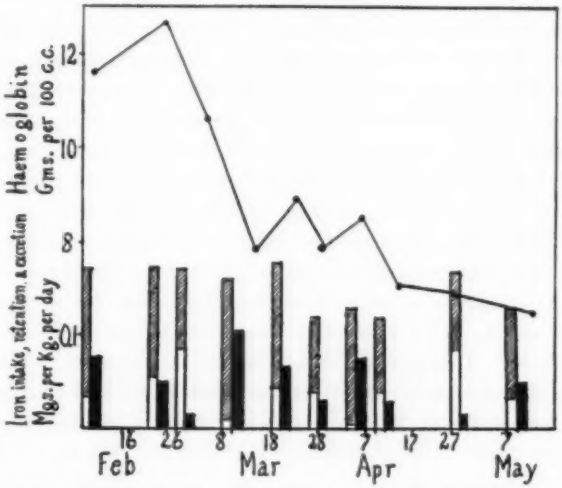

CHART 5. IRON RETENTION AND EXCRETION IN AN INFANT DURING DEVELOPMENT OF ANEMIA (CASE 21)

For explanation of symbols see previous chart

at variance with the results of Barer and Fowler (24) who found a negative iron balance in anemic adults, but a positive balance in non-anemic controls. The difference in results at the two ages indicates a possible fundamental difference in the behavior of the tissues toward iron, and suggests the possibility that in infancy there is an increased "avidity" of the "stores" for iron which might conceivably compete with the hemoglobin for the available iron. In the next paper it will be shown that ingested iron is stored in the tissues before being utilized for hemoglobin formation, so that utilization is not

merely a matter of a supply of available iron. In some cases at least, one may have to consider the possibility that the "avidity" of the tissues for iron, may be a factor in the development and continuance of anemia.

COMMENT

It is evident from the data presented in an earlier paper, and from the work reported here, that no basis has been found for the usually accepted notion of iron deficiency. The old idea that goes back to Bunge, that milk is totally inadequate as a source of iron for hemoglobin formation, is true only for the period of rapid growth, when the stores are being drawn on (1). In an extensive study, no instance has been found of a loss of iron from the body which could be considered of any significance in the development of iron deficiency. Only in the case of the premature baby or twin, in whom birth stores are low in relation to the later gain in weight, may iron deficiency, in its usually accepted sense, play an important part in producing anemia.

There is, however, another conception of iron deficiency which is applicable to cases of anemia other than those occurring in premature babies or twins. This conception contains the idea of lack of availability of iron. It is possible that iron may be deposited in the tissues in a form that renders it almost as unavailable as if it were outside the body. The fact that for the first six months of life the iron of the tissues is readily utilized when needed, makes this hypothesis less tenable. The alternative is that iron is not utilized because the conditions are not right for its utilization. In other words, after anemia has developed for various reasons, iron is not utilized but is stored to be utilized when the conditions are right. The effect of medicinal iron is, on the one hand, to supply iron in an available form, and, on the other, to alter the conditions by stimulation or otherwise, so that hemoglobin may again be formed. An attempt to study the action of medicinal iron is the subject of the next paper of this series.

SUMMARY

Iron balance was studied in a large number of infants for the purpose of ascertaining the factors influencing retention of iron on milk and cereal diets.

There was evidence to show that, on the average, approximately 60 per cent of the iron taken in with the food, was absorbed. Excretion tended to vary with age, being somewhat greater in the first three months of life, but thereafter relatively constant.

Retention of iron tended to be below the average normal during infections, and in infants not receiving vitamin D. In cases of infection the diminished retention seemed to be due to the reduction in intake caused by the loss of appetite; while in cases of vitamin D deficiency there appeared to be an actual diminution of iron absorption. Deviation from the normal retention, however, was never enough to contribute significantly to iron deficiency.

Retention of iron tended to be above the average in cases of anemia even when the hemoglobin was falling. While the amount of extra retention was not great, it was felt that this occurrence rather opposed the idea of iron deficiency in the ordinarily accepted sense, and pointed to the possibility that "avidity" of the tissues for iron might be a factor in the production of anemia. It was pointed out that the conception of iron deficiency must include the idea of availability of iron.

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IRON METABOLISM IN INFANCY

II. The Retention and Utilization of Medicinal Iron[1]

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This paper is the third of three dealing with iron metabolism in infancy. The first paper (1) reported an attempt to study the quantitative aspects of iron deficiency, to find out under what conditions there might occur an actual lack of iron in the body. It was found that although there was likely to be a negative balance in the first two months of life, the loss of iron during this period was of no importance. Moreover, it was found that at no time was there a deficiency of iron in the body, sufficient to affect the level of hemoglobin in the first six months of life, though it was calculated that by this time the stores would probably have been exhausted. It had previously been determined in rats (2) that anemia occurring on milk diets was essentially due to a gain in weight, since the iron of the body remained relatively constant and with it the total calculated hemoglobin. In other words, the anemia depended on the dilution of a constant amount of hemoglobin iron in the growing body; and anemia developed rapidly in the rat because the rat grew relatively rapidly.

With this observation as a basis, it was calculated that iron deficiency was virtually impossible in the human infant except in cases in which there was either (1) a rapid gain in weight in relation to stores at birth, or (2) a significant loss of iron from the body. The first might occur in premature babies, and in babies whose stores at birth were deficient as in offspring of anemic mothers. The second has not been found to occur in any case thus far studied, but would, of course, be present in cases with hemorrhage that had not received

[1] The expenses of this investigation were met by a grant from the Howland Memorial Fund.

transfusion. Even in cases with defective storage, the relative weight gain would be insufficient to account for anemia of the severity often found in so-called nutritional anemia. Lichtenstein's (3) figures showing a negative iron balance in premature babies have not been confirmed either by Snelling (4) or by my own figures in the preceding article (5).

In the second paper (5), the figures for iron retention on ordinary milk and cereal diets were analyzed to determine, if possible, the conditions influencing the retention of iron. With the exception of a lower iron intake and, to a minor extent, age, nothing was found to interfere with absorption or to increase excretion sufficiently to be a factor in producing iron deficiency. As a result of earlier work it had been thought possible that infection might cause a significant loss of iron. But this hypothesis was not confirmed.

As a result of these studies showing the improbability of a quantitative iron deficiency as a cause of nutritional anemia in infancy it was necessary to turn to a study of the internal metabolism of iron. Two possibilities offer themselves: (1) that iron is deposited in the tissues in relatively unavailable form; (2) that the anemia is due to conditions within the body which interfere with the formation of hemoglobin. Medicinal iron may accordingly either (1) supply iron in a form more available for utilization, or (2) cause a change in conditions leading to a more rapid hemoglobin formation. It was to decide between these possibilities that the work reported in this paper was undertaken.

The procedure was as follows: The infant to be investigated was restrained on a metabolism frame for the collection of uncontaminated stools. Sometimes he was studied for a preliminary period, during which he was kept on a low iron, milk and cereal feeding, but usually, especially in the later part of the work, this was omitted to save time. Then, without being removed from the frame, he was given ferric ammonium citrate in his feeding over a period of 4, 5 or 6 days. In the earlier work a period continuous with the "iron" period and immediately following it, was included to take care of the supplementary excretion of iron. Later, when it was found that all the iron that was going to be excreted appeared in the stools the day following the last dose of ferric ammonium citrate, this supplementary period was

TABLE I

Data and calculated figures in infants up to six months of age

CASE NUMBER	AGE	WEIGHT	INITIAL HEMOGLOBIN	DURATION OF "IRON" PERIOD	RETICULOCYTE RESPONSE	LATENT PERIOD	DURATION OF RISE IN Hgb.	IRON, MG. PER KG. PER DAY		IRON, MG. PER KG. TOTAL FOR PERIOD			IRON MG. PER KG. PER DAY RATE OF UTILIZATION	REMARKS
								Intake	Retention	Retention	Utilization	Storage		
				days		*days*	*days*							
14	1 m.	2.8	9.7	5	0	6	7	9.7	−.43	−2.1	5.0	−7.1	.7	See chart 5
7	6 w.	2.5	14.5	5	0	—	—	6.9	2.6	12.8	−7.2	20.0	−.55	
17	6 w.	4.3	15.7	4	0	—	—	3.5	.46	1.8	−1.8	3.7	−.18	No effect of iron whatever
15	6 w.	3.6	12.6	5	0	7	5	8.6	−.3	−1.7	4.2	−5.9	.84	"Stimulation" or coincidence
7	2 m.	3.0	10.6	5	0	4	12	5.1	1.5	7.4	7.7	−.3	.64	
16	2 m.	4.4	13.2	5	0	5	9	4.2	.7	3.5	5.5	−2.0	.61	
10	2½ m.	3.5	10.9	5	—	0	12	11.1	2.6	13.1	4.0	+9.1	.33	
9	3 m.	2.8	11.5	6	—	0	5	4.6	1.6	9.6	7.1	2.5	1.01	
8	3 m.	2.2	5.5	6	—	0	12	4.9	2.2	13.2	−2.3	15.5	−.19	
12	3½ m.	5.5	12.0	6	—	0	12+	3.6	.27	1.6	6.0	−4.4	.5	"Stimulating" action of iron (see ch. 9)
12	4 m.	6.6	13.1	5	—	0	12	2.3	.24	1.2	5.0	−3.8	.42	"Stimulating" action of iron (see ch. 9)
36	4 m.	4.6	10.4	5	—	0	10	8.3	4.7	23.7	.65	23.0	.06	
11	4 m.	6.7	13.7	6	—	0	12	2.7	−.12	−.75	3.7	−4.5	.31	Good "stimulation" in spite of no retent
12	5 m.	7.4	12.6	5	—	—	—	2.5	.08	.41	.82	−.41	.05	See chart 9
17	5 m.	7.2	13.4	5	—	3	9	5.3	.75	3.8	6.8	−3.0	.75	"Stimulating" action of iron
22	6 m.	5.3	12−	5	0	0	14	7.7	.91	4.5	3.6	.95	.26	

To save space the data are presented in the form in which they are used in constructing the charts, *e.g.*, the actual mg. of iron given during the period are omitted, and intake is reported as daily intake per kg. of body weight. The amounts of feeding given, the iron content of the stools, and the urinary excretion have likewise been omitted from the table.

In the case of infants under six months of age, the initial fall and subsequent rise in hemoglobin were independent of the amount of iron given, although in some cases the iron appeared to stimulate hemoglobin formation (see Chart 4).

TABLE II

Data and calculated figures in infants of six months to one year

CASE NUMBER	AGE	WEIGHT	INITIAL HGB.	DURATION OF "IRON" PERIOD	RETICULOCYTE RESPONSE	LATENT PERIOD	DURATION OF RISE IN HGB.	IRON, MG. PER KG. PER DAY		IRON, MG. PER KG. TOTAL FOR PERIOD			MG. PER KG. PER DAY RATE OF UTILIZATION	PERCENTAGE OF RETAINED IRON UTILIZED	REMARKS
								Intake	Retention	Retention	Utilization	Storage			
				days		*days*	*days*								
10	6½ m.	6.3	11.2	5	±	5	8	5.2	.86	4.3	3.95	.35	.49	92	
33	7½ m.	6.8	13.4	6	0	—	—	3.3	.075	.45	.3	.15	0	—	
29	8 m.	5.3	6.5	5	+	0	14	5.9	.59	3.0	2.45	.55	.175	82	
29	8 m.	5.8	6.9	5	++	5	9	12.4	3.3	16.5	7.9	8.6	.79	47	
35	8 m.	5.4	8.5	5	0	—	20?	7.5	2.8	13.9	1.5	12.4	.08	11	
12	8½ m.	10.5	10.5	5	±	—	18?	4.1	.96	4.8	2.0	2.8	.11	42	
30	8½ m.	6.9	5.8	5	+	—	11?	4.8	.64	3.2	.58	2.6	.05	18	
30	9 m.	7.4	5.6	5	+	10	12	4.7	1.1	5.5	7.5	−2.0	.62	136	Copper
30	9 m.	7.7	7.1	5	0	0	14	4.3	2.2	11.0	4.5	6.5	.32	41	
21	8½ m.	6.2	6.5	5	+	7	7	4.6	1.5	7.5	1.6	5.9	.23	23	
21	9 m.	6.5	6.9	5	0	—	12?	8.0	0	0	1.0	−1.0	.08	—	
34	9 m.	6.0	9.7	5	±	—	—	2.6	.17	.85	0	.85	0	0	
34	10 m.	6.8	8.9	5	±	5	13	4.9	.56	2.8	.59	2.2	.045	21	
27	10 m.	8.0	8.1	5	±	5	10	3.4	.35	1.75	1.0	.75	.10	57	
31	10 m.	9.1	7.2	5	±	2	9	3.5	.96	4.8	3.4	1.4	.38	71	
32	11 m.	6.7	6.9	5	±	6	7	2.6	.03	.14	4.2	−4.05	.6	300	"Stimulating" action of iron
32	11 m.	7.1	8.1	5	0	—	12?	4.9	.28	1.4	−.3	1.7	−.023	−20	

The reticulocyte rise is represented by symbols rather than by actual figures merely for convenience in reading the tables.

The latent period is the period that elapses between the beginning of the "iron" feeding and the beginning rise in haemoglobin. 0 means that there was no latent period, the rise in hemoglobin being evident at the time of the first determination after iron was given. A dash means either that there was no hemoglobin rise or that the first determination after beginning iron therapy was performed too late.

The duration of the rise in haemoglobin was measured from the end of the latent period to the point at which the hemoglobin rise came to an end, as described in the text.

In these cases and those in Table III there is a much closer relation between iron retention and utilization than in those recorded in Table I. Case 32 is an exception (2 periods) and also possibly case 10.

omitted, the iron intake being continued for five days and the collection of stools for six. In this work the iron was added each morning to the day's supply of milk and an aliquot of each day's feeding was taken. The aliquots were mixed at the end of each period and the amount of iron actually ingested was determined by analysis. Strict account was kept of refusal and vomiting. The treatment of the stools and urine and the analysis of iron have been given in detail in previous papers (1, 5) and need not be repeated here.

TABLE III

Data and calculated figures in infants over one year of age

CASE NUMBER	AGE	WEIGHT	INITIAL HGB.	DURATION OF "IRON" PERIOD	RETICULOCYTE RESPONSE	LATENT PERIOD	DURATION OF RISE IN HGB.	IRON MG. PER KG. PER DAY		IRON MG. PER KG. TOTAL FOR PERIOD			RATE OF UTILIZATION MG. PER KG. PER DAY	PERCENT OF RETAINED IRON UTILIZED	REMARKS
								Intake	Retention	Retention	Utilization	Storage			
				days		*days*	*days*								
37	12½ m.	9.0	9.6	5	0	—	—	3.0	.71	3.6	0	3.6	0	0	
28	14 m.	9.3	3.5	5	++	3	6	3.2	.86	4.3	4.7	−.4	.78	112	
28	14½ m.	10.1	4.5	5	±	5	4	2.9	.76	3.8	2.1	1.7	.52	54	
28	15 m.	10.5	5.1	5	+	5	9	3.1	1.2	6.1	5.8	.3	.64	95	Copper
28	15½ m.	11.0	6.4	5	++	7	9	3.0	1.26	6.3	6.3	0	.70	100	Copper
23	15 m.	8.0	4.3	5	++	5	9	7.9	2.0	10.2	5.0	5.2	.56	49	
38	15 m.	8.0	6.5	5	−	0	8	3.6	.76	3.8	2.8	1.0	.35	74	
26	18 m.	8.1	5.9	5	+	4	12	3.1	.72	3.6	5.1	−1.5	.42	142	Copper
26	18 m.	8.9	6.9	5	+	0	20	3.4	.41	2.0	2.4	−.4	.12	120	
26	18 m.	8.7	7.9	5	±	0	5	5.2	.96	4.8	3.1	1.7	.62	65	
25	17 m.	8.1	8.5	5	0	4	9	4.0	1.02	5.1	4.3	.8	.48	85	
24	19 m.	8.4	5.5	5	+	0	17	3.2	1.18	5.9	3.2	2.7	.19	55	
24	19 m.	8.6	6.9	5	+	5	13	4.0	1.8	8.8	3.2	5.6	.25	36	
24	19 m.	9.2	7.6	5	++	5	12	3.2	.71	3.5	4.8	−1.3	.40	137	Copper

The amounts of iron used were small in comparison with the usual therapeutic dose. This was partly for the purpose of giving available iron and at the same time avoiding maximum stimulation, and partly to learn what the minimum effective dose was.

It had been my purpose to make this study as complete as possible. Circumstances forced the abandonment of the problem before it was finished, and the incompleteness of the data will be evident. At this point it may be said that the study lacks data on iron intake at a low

and at a high level; it contains too few cases of severe anemia, and too few non-anemic cases above nine months of age. As it stands, conclusions can be no more than tentative, but even with this reservation the study throws some light on the nature of "iron-deficiency" anemia.

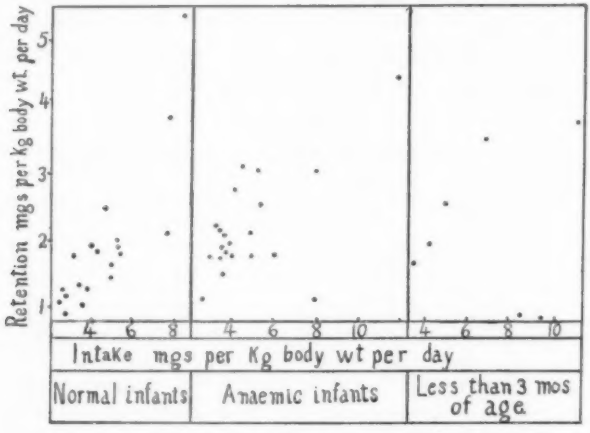

CHART 1. RELATION OF IRON RETENTION TO INTAKE

Anemic infants include those whose hemoglobin at the beginning of the period of observation was below 8 gm. per 100 cc. Those with mild anemia are therefore included with the normal infants. No attempt has been made to average these points but one has the impression that the curve rises more steeply at lower levels of intake in the anemic group than in the non-anemic.

RESULTS

The data, presented in Tables I, II and III, will be considered under four headings: (1) Retention of iron; (2) Effect of iron in stimulating blood formation; (3) Utilization of iron for hemoglobin formation; (4) Storage of iron.

1. Retention of Iron

The relation of retention to intake (Chart 1). Infants from birth to three months show such wide variations in retention of iron that it seems best to disregard them in the discussion of this problem. In infants over 3 months of age, there appears to be a definite correlation

between intake and retention when these are expressed in mg. per kg. of body weight per day (see chart). Figures from the literature bear out this correlation. Several statements have, however, been made that iron equilibrium is always attained whatever the dose of iron. Lintzel (6), who is responsible for this statement, and Reimann, Fritsch and Schick (7), who confirmed it, did not administer iron in amounts of more than 1 to 2 mg. per kg. of body weight per day; such intakes correspond to that portion of the chart where equilibrium is to be expected. Their work, which has been widely quoted, gives, therefore, an entirely erroneous impression.

In the second place, anemic infants appear to retain iron better than the non-anemic. There should be more figures to make this certain, but it is a phenomenon which has also been observed by others (7, 8).

In the third place, the figures converge to a point which indicates that to attain equilibrium 2.0 mg. of iron per kg. of body weight per day must be given. What happens when the intake is below this level is not clear. In the previous paper it was shown that, in the case of food iron, equilibrium was attained at an intake of about 0.15 mg. per kg. per day and that above this level the balance became increasingly positive. We have, then, the curious possibility that when small amounts of iron are added to a diet on which there is a positive balance, the balance may become negative. This was actually the case in the man studied by Resnikoff, Toscani and Fullerton (9); and Müller (10) also found that medicinal iron might lead to a negative balance. Müller, however, made use of Neumann's method for iron analysis which is not accurate; and other figures in the literature make it probable that on an intake below 2.0 mg. per kg. per day the body is virtually in equilibrium.

Relation of retention to absorption. It is impossible to draw any conclusions regarding the relation of absorption to intake, on the basis of the few data at hand. The question here is whether all the absorbed iron is represented by the retention or whether some of the absorbed iron is later reexcreted. If reexcretion occurs, it must be extremely rapid as is indicated by the data in Tables IV and V. In Table IV it may be seen that in the period immediately following that in which the added iron was given, the negative balance was, with one excep-

TABLE IV

The Lag in the Output of Iron

CASE NUMBER	DATE	TOTAL INTAKE	TOTAL OUTPUT	AVERAGE OUTPUT PER DAY DURING "IRON" PERIOD	LAG IN OUTPUT	
		mg.	mg.	mg.	mg.	
9	12/ 9–15	78	46	7.7	5.4	
	12/15–18	1.7	7.1			
8	12/ 9–15	65	33	5.5	3.2	
	12/15–18	1.4	4.6			
36	2/12–17	191	75	15	7	
	2/17–22	7	14			
10	2/ 8–13	195	85	17	63	No stool on last day of "high iron" period
	2/13–18	4.7	68.3			
11	1/ 9–15	107	99	20	13	
	1/15–18	2.7	16			
12	1/ 9–15	117	100	16.5	8	
	1/15–18	2.6	10.8			
12	2/ 5–10	75	60	12	7	
	2/10–15	6.7	13.5			
12	2/24–3/1	92	71	14	17	
	3/ 1–6	9	26			
33	1/ 9–15	133	113	19	17.5	
	1/15–18	2.9	20.4			
34	4/14–19	77.5	55	11	17	
	4/19–24	4.2	21.2			
34	5/ 8–13	165	137	27	9	
	5/13–17	5.6	14.5			
35	3/15–20	202	107	21	19.5	
	3/20–25	4.2	23.8			
22	3/16–21	202	148	30	29	
		4.2	33			
16	3/14–19	90	69	14	5.5	
	3/19–24	7.2	12.7			
17	3/ 3–7	60	43	11	8.5	
		3.8	12.4			

This table is intended to demonstrate the amount of lag in iron output after the high iron intake is discontinued. In these cases the "after period" (lower line) is continuous with the "high iron" period (upper line). In the last column the figures are obtained by subtracting the intake from the output in the "after period" and are intended to represent the iron that continues to appear in the stools after the end of the "high iron" period, on the assumption that the infant would otherwise be essentially in iron balance. These figures should be compared with those in the preceding column representing average daily output during the "high iron" period. The figures in the latter column should actually be somewhat higher than represented, because in calculating them no account was taken of the lag in the first appearance of the "high iron" in the stools. This "error" will not, however, make any difference in the interpretation. From the fact that the figures in the last column are usually smaller than those in the preceding column, it may be deduced that the lag in iron output is probably less than twenty-four hours—about the time required for the passage of material from the mouth to the anus. It is likely, then, that at the level of iron intake employed in these cases retention may be considered equal to absorption, and that little of the iron taken into the body is reexcreted.

tion, no more than the average daily output of the "high iron" period. This suggested that all the iron that was going to be eliminated would have appeared in the stools within thirty-six hours of the last ingestion of added iron. Table V shows this to be the case, for it is clear that if one extra day was allowed for the collection of stools representing

TABLE V

CASE NUMBER	INTAKE		OUTPUT		MG. PER DAY IN PERIOD FOLLOWING HIGH IRON	
	Date	Mg. iron	Date	Mg. iron	Intake	Output
28	1/15–20	147	1/15–21	108	1.92	1.23
	1/20–24	7.68	1/21–24	3.68		
15	5/ 4–9	154	5/ 4–10	160	0.81	1.07
	5/ 9–13	3.24	5/10–13	3.22		
14	12/15–20	134	12/15–21	140	0.696	0.85
	12/20–25	3.48	12/21–25	3.41		
17	6/10–15	191	6/10–16	164	1.76	1.47
	6/15–19	7.05	6/16–19	4.41		
10	6/10–15	164	6/10–16	137	1.51	1.36
	6/15–19	6.04	6/16–19	4.08		
12	6/11–16	211	6/11–17	161	2.01	3.5
	6/16–20	8.06	6/17–20	10.6		

This table confirms the conclusion reached from the figures in the preceding table, namely, that the lag in iron output after the end of the "high iron" period is not more than a day. It is evident that if one extra day is allowed for the collection of stools to represent the output of the "high iron" period, the child thereafter is essentially in iron balance. It is interesting to note that of the three cases with negative balance in the "after" period two had a negative balance during the "high iron" period and one had been given a relatively high intake. In Table IV, of those cases in which the lag in output exceeded or nearly equalled the daily output, cases 12, 33 and 34 were very nearly in iron balance during the "high iron" period while cases 35 and 22 were given relatively high intakes. One may infer, then, that the "avidity" of the tissues for iron is an important factor in the retention of iron, but that when high iron dosage is employed this "avidity" may be exceeded, resulting in an increase in the excretion lag, i.e. continued output of an excess of iron in the first day or two following discontinuance of the "high iron" period.

the "high iron" period, then, in the days immediately following, the infant was in iron equilibrium. This was true up to about 50 mg. per day intake. It is possible that at higher levels of intake the lag in output might be greater. Both Lintzel (6) and Reimann, Fritsch and Schick (7) found essentially the same thing, that the iron which was retained before equilibrium was established on their "high iron"

diets, was reexcreted only in small part; and that within three days of the withdrawal of medicinal iron the body was in iron equilibrium. They, however, as has been pointed out, used low (per kg.) intakes. Hutchison, using higher intakes, also found that "reexcretion" of iron was limited to the first period following the cessation of "high iron."

Relation of retention to initial hemoglobin level. It has already been pointed out that retention appears to be greater in the anemic than

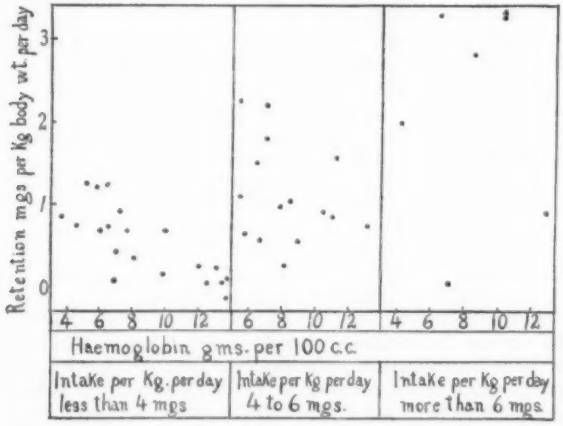
Retention mgs per Kg body wt. per day
3
2
1
0
4 6 8 10 12
6 8 10 12
4 6 8 10 12
Haemoglobin gms. per 100 c.c.
Intake per Kg. per day less than 4 mgs
Intake per Kg per day 4 to 6 mgs.
Intake per Kg per day more than 6 mgs.

CHART 2. RELATION OF INITIAL HEMOGLOBIN LEVEL TO IRON RETENTION IN INFANTS OVER THREE MONTHS OF AGE

It may be noted that the correlation is best expressed by the lower levels of intake while at intakes above 6 mg. there is no apparent correlation. This fact may explain disagreements in the literature, as discussed in the text.

in the non-anemic individual. This statement, however, applies more particularly to daily intakes below 6 mg. per kg. of body weight. In Chart 2 the correlation between retention and initial hemoglobin is shown. Barer and Fowler (11, 12, 13, 14) in their extensive study of iron metabolism, did not find such a correlation. In my cases there was a lag of about four to eight days between the beginning of the "high iron" period and the beginning of a rise in hemoglobin. Since the period during which iron was given lasted only five or six

days, it follows that the retained iron must all have been first deposited in the tissues and utilized later. It is, then, the "avidity" of the tissues for iron rather than the hemoglobin level *per se* that would be expected to influence retention. It seems reasonable to suppose that it is in the "avidity" of the tissues for iron that the cases reported here differ from those of Barer and Fowler. The factors influencing the "avidity" of the tissues for iron are not known, but it is not unlikely that age, type of anemia, and the relative level of tissue iron may play a part. The experiments of Fontès and Thivole (15) showing a progressive increase in iron retention in dogs depleted of iron by successive large bleedings have an important bearing on this question. It was pointed out in the previous paper that all of Barer and Fowler's cases with anemia had a considerably increased iron excretion in the periods before iron was given, whereas in the cases reported here the tendency was to low excretions. (See previous paper (5).)

Relation of retention to age. Age appeared to have little influence on retention except that under the age of three months, retention bore no apparent relation to intake. There was only one case of anemia (No. 8) in this age group, and in this patient there was good retention, even though the anemia was dependent on increased hemolysis which would not be expected to cause low levels of tissue iron. A twin brother of this child (No. 9) with normal hemoglobin on the same iron intake, retained almost exactly the same amount.

Copper was without influence on retention. The lack of influence by copper was found also by Schlutz, Morse and Oldham (16) and by Maurer, Greengard, Curtis and Klüver (17).

2. *Effect of Iron in Stimulating Blood Formation*

This is an important aspect in the study of the effect of medicinal iron about which there has been much controversy. It can hardly be doubted by those who have had experience with the treatment of anemia that the giving of iron does more than merely supply a necessary building stone for hemoglobin. One of the most striking effects in infants is the improvement in appetite and in general well-being and disposition, which occurs before there is any rise in hemoglobin worth considering. The most obvious objective evidence of improvement is, however, the rise in reticulocytes, which is so striking as to

be used as the means for determining the optimum dose of iron. Whether the rise in reticulocytes is to be considered evidence of "stimulation" of red blood cell formation or merely of a more rapid "maturation" of the cells depends upon the definition of these two terms. There can, however, be no doubt that the administration of iron in anemia is followed by a more normal course of blood formation,

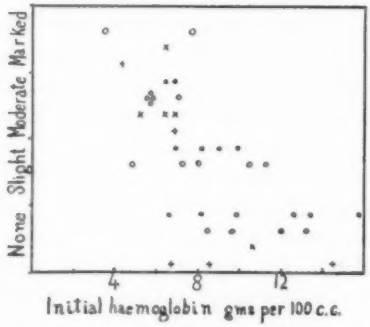

CHART 3. RELATION OF RETICULOCYTE RISE TO INITIAL HEMOGLOBIN LEVEL

The solid circles represent total period retentions below 3 mg. per kg. of body weight, the hollow circles from 3 to 6 mg., the diagonal crosses from 6 to 10 mg., and the vertical crosses above 10 mg.

As may be seen, initial hemoglobin has a pronounced influence on reticulocyte response, while iron intake and retention has none. As explained in the text, there are probably other factors having an effect on reticulocyte response that are at present unknown and therefore uncontrollable. Of course, the presence of an infection is one such factor not, however, present in any of these cases.

It must not be thought that reticulocyte percentage gives a quantitative measure of that rather elusive concept "stimulation." The reticulocyte response is one of the phenomena associated with "stimulation" and appears to measure with some degree of accuracy the relative rate at which red cells are brought to a sufficient maturity to enter the blood stream.

and that the evidence for the change to normal is found in the reticulocyte rise.

It was, therefore, a matter of surprise that the only factor that could be found to have a demonstrable influence on the reticulocyte rise was the initial hemoglobin level (chart 3). Neither the intake nor the retention of iron had any influence, and yet we know from clinical

experience that to obtain maximum responses large doses of iron must be given. In fact, the necessity of giving large doses to obtain maximum response is the reason for giving large doses in clinical work. It seems quite possible that other factors, at present little understood, have a dominating influence upon the reticulocyte response when the dose of iron is relatively small, so that whatever effect iron may have, will not be apparent unless these other factors can be recognized and controlled.

3. Utilization of Iron for Hemoglobin Formation

The determination of utilization requires the use of calculations that rest on certain assumptions: (1) that blood volume bears a fairly constant relation to body weight (76 cc. per kg.); (2) that the total hemoglobin of the body can be calculated approximately from the blood volume and the percentage of hemoglobin in the blood; (3) that the iron of hemoglobin is constant (0.34 per cent). All three assumptions are subject to possible revision, but for practical present purposes they may be accepted as true. The total hemoglobin iron has been calculated from the data at the time iron therapy was started, and again at the end of the period of hemoglobin rise, usually two to three weeks after the commencement of the "high iron" intake.[2] The difference between the calculated total hemoglobin iron at the beginning and at the end of the period of hemoglobin rise is considered to represent the utilization of iron.

Relation of utilization to age. The chart showing utilization in relation to age in non-anemic patients (hemoglobin above 8.0 g. per 100 cc.) has been omitted. It showed no more than we already know concerning the behavior of the hemoglobin in the first six months of life. It is interesting, however, that in the slightly anemic patients the retention of iron bore no relation to utilization, although a stimulating action could be demonstrated in a number of instances (chart 4, also cases 17, 15 and 11). In other words, the low hemoglobin levels of the second half-year of life cannot be due solely to deficiency of iron, if the level of hemoglobin is uninfluenced by the known pres-

[2] The actual point considered to be the end of the hemoglobin rise was the earliest point at which the hemoglobin curve definitely levelled off or actually began to decline after reaching a maximum.

ence of available iron (as in cases 35, 12 and 37, in which there was no rise in hemoglobin in spite of good retention and relatively low initial hemoglobin).

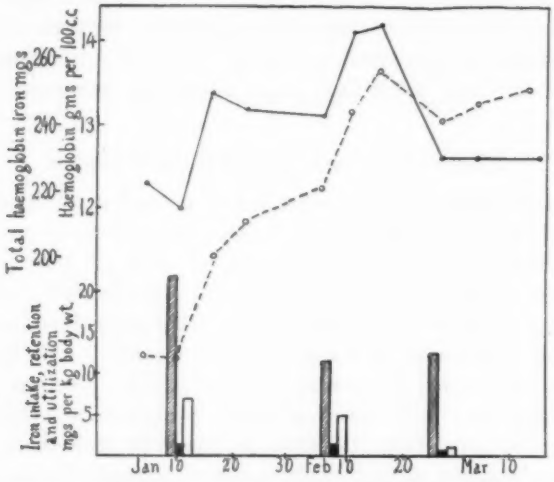

CHART 4. HEMOGLOBIN PER CENT AND TOTAL HEMOGLOBIN IRON IN CASE 12 FOLLOWING "HIGH IRON" PERIODS

The solid line represents hemoglobin percentage; the broken line represents total hemoglobin iron calculated from probable blood volume and hemoglobin per cent. The shaded column represents iron intake; the solid column, iron retention; the outlined column, iron utilization.

It is evident that the ingestion of iron was followed on two occasions by utilization of an amount of iron that far exceeded the amount retained. This is an example of what might be called the "stimulating" action of iron, that is particularly to be seen at this time of life. It lasted about ten days in two instances and was absent on the third occasion. The net result of the periods of "stimulation" was nil. It is quite probable that the level of hemoglobin was determined by conditions inside the baby and since these conditions were relatively good to start with, the effect of iron was seen only during the period of stimulation, and no permanent change was effected. At this time of life hemoglobin response appears to bear little relation to the amount of iron retained.

Relation between utilization and retention. Chart 6 represents the relation between utilization and retention of iron in infants over six months of age. The cases under six months were excluded because

the utilization of iron was uninfluenced by retention in the majority. The effect of copper is well shown in this chart. Leaving out these cases, there appears to be a fairly good correlation between retention and utilization. This is at variance with the finding of Fowler and

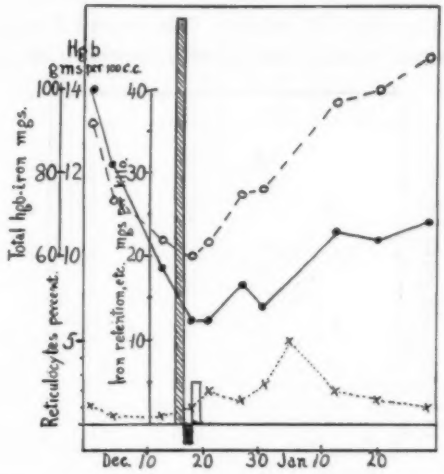

CHART 5. HEMOGLOBIN PER CENT AND TOTAL HEMOGLOBIN IRON IN CASE 14 (FOUR WEEKS OF AGE)

The solid line represents hemoglobin concentration; the broken line, total hemoglobin iron calculated from probable blood volume and hemoglobin percentage; the dotted line, reticulocyte percentage. The shaded column represents iron intake; the solid column iron retention; the outlined column, utilization.

At this age it is impossible to exclude coincidence since spontaneous reticulocyte rise may occur, but it is not unlikely that the medicinal iron may have "stimulated" the rise in hemoglobin before it would have occurred under ordinary conditions. The lack of any relation between retention and utilization is evident. The presence of possible stimulation in the presence of negative retention raises the question of the relation of absorption to retention. There are other similar cases with negative retention in which the "stimulating" action is hardly to be doubted.

Barer that there is no such correlation. Their total iron intake over a long period was considerably larger than the amount given in the cases reported here; and it is probable that with even the lowest retention exceeding the need for hemoglobin formation, the differences

in retention would not be expected to have much influence on utilization. Folkman and Ullrich (18) found that there was more iron retained in cases in which the hemoglobin rose than in those in which it did not rise.

Influence of copper upon utilization. This is clearly shown in chart 6, confirming the effect in animals. Barer and Fowler, as well as Maurer, Greengard, Curtis and Kluver, have also demonstrated the

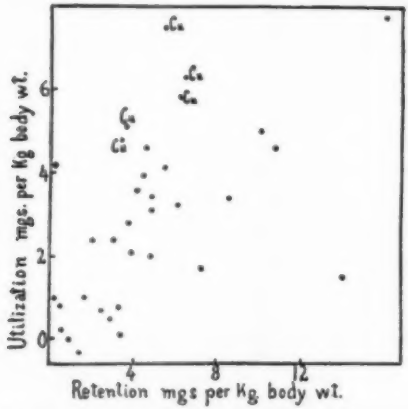

CHART 6. RELATION OF RETENTION TO UTILIZATION OF IRON

None of the cases in Table I are included in this chart, because of the lack of any correlation between retention and utilization in the majority of infants under six months of age. Of course, in individual cases there may be some apparent relation, but the group must be included or excluded as a whole. In the infants over six months the cases that do not follow the general trend are exceptional. (The dots marked Cu represent cases in which copper was given along with the iron.)

effect of copper in increasing the utilization of iron. When utilization is charted, not as the total amount of iron used in hemoglobin formation, but as the daily amount so used (i.e. rate of utilization) the effect of copper is much less easy to detect. This may have some significance in indicating that the effect of copper is to make iron more available rather than to catalyze the formation of hemoglobin. The number of cases is too small to be more than suggestive on this point.

Influence of initial hemoglobin level upon utilization. The correla-

tion of rate of utilization with initial hemoglobin shown in chart 7, is not as clearly demonstrated as might have been expected. Little relation was found between utilization of iron and the rise in reticulocytes. This rather unexpected result will be discussed later.

4. *Storage of Iron*

In general the period of "high iron" feeding was over before the hemoglobin began to rise, so that it is obvious that the iron is first

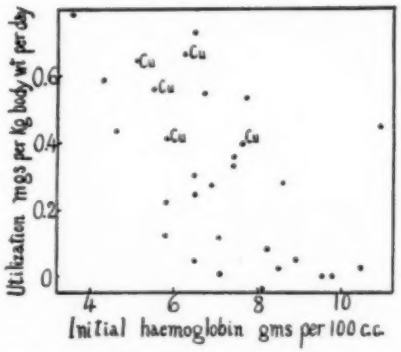

CHART 7. RELATION OF RATE OF UTILIZATION OF IRON TO THE INITIAL HEMOGLOBIN LEVEL

As may be seen, there is a correlation, though it is not very close. In all work with iron metabolism the scattering of the figures precludes good correlation without the use of many more figures than are included in this chart.

It is interesting that the effect of Cu is not demonstrated in this chart, as it is in charts 6 and 8. Copper appears to have more influence on total utilization than on the rate of utilization.

deposited in the tissues and then later utilized for hemoglobin formation. The problem of utilization is then intimately associated with that of availability of stores.[3] Since all the retained iron is first stored, the percentage of retained iron that is utilized can be used as a measure of the tendency to withdraw iron from the stores.

In chart 8 the percentage utilization is charted against retention. It is evident that in the anemic cases utilization is much more efficient

[3] It must be realized that "storage" is used here in a broad sense, as including that portion of the retained iron not being utilized for hemoglobin formation.

at lower levels of retention, at 2[illegible] mg. per kg. of body weight and lower amounting to 100 per cent or even more. The more efficient utilization at lower retentions is also shown by Brock and Hunter's, and by Hutchinson's figures (8). This has a bearing on one error of interpretation that has recently crept into the literature on iron metabolism.

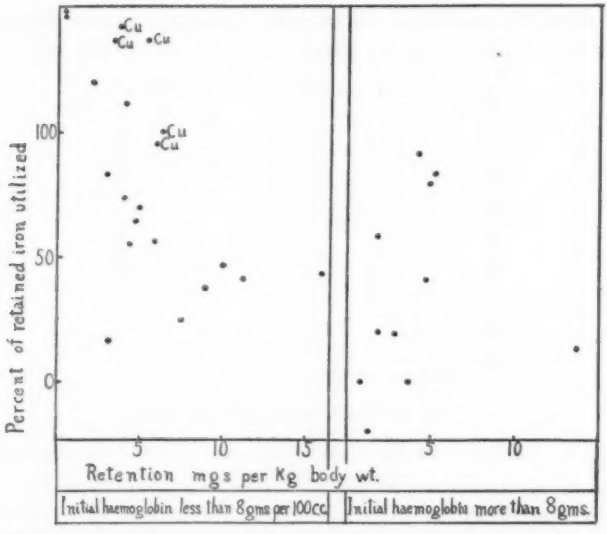

CHART 8. RELATION OF PERCENTAGE UTILIZATION TO IRON RETENTION AND THE INITIAL HEMOGLOBIN LEVEL

In spite of the small number of observations it is clear in the anemic cases that iron is utilized more completely at lower levels of retention than at higher levels; while in the non-anemic cases there seems to be no correlation. Utilization above 100 per cent of course means that iron is drawn from the stores for hemoglobin formation.

Heath, Strauss and Castle (19) found that when iron was injected intramuscularly it was practically all utilized in hemoglobin formation, and from this it has been inferred that when iron is given by mouth its utilization for hemoglobin is a measure of its retention (20, 21). A second inference has also been made, that the large doses required in clinical work are necessary in order to give a sufficient retention for

maximum hemoglobin formation. The results reported here, as well as the work on iron balance in the literature, give ample proof of the

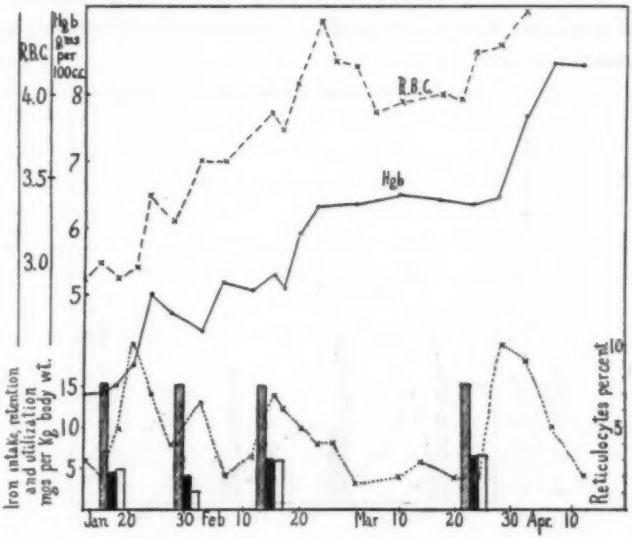

CHART 9. HEMOGLOBIN, RED CELLS AND RETICULOCYTES IN A CASE OF SEVERE NUTRITIONAL ANEMIA (CASE 28) AFTER MEDICINAL IRON

This case is one of a rather severe nutritional anemia with typical history of prolonged milk diet and few infections. The dosage of iron, 3 mg. per kg. per day, was less than one-tenth of the usual therapeutic dose. Yet there was an adequate reticulocyte response, if not quite maximum. It is one in which the behavior toward iron therapy is what one would expect from a case with little or no available iron to draw on. Practically all the retained iron is utilized except in the second period, so that the limit of each rise in hemoglobin appears to be set by the amount retained. It may be noted, however, that the end of the rise in hemoglobin also is more or less coincident with the termination of the reticulocyte response. The relatively small rise in the second period is coincident with a relatively slight reticulocyte response, and, in this instance, only about one-half the retained iron was utilized. This chart should be compared with the next one. (See chart 5 for meaning of symbols.)

falseness of these inferences. The amount of iron injected by Heath, Strauss and Castle was small, less than 1 mg. per kg. and therefore

would correspond to a level of retention at which utilization would be expected to amount to 100 per cent or more. Heilmeyer (22) injected only 10 mg. of iron in combination with ascorbic acid and obtained a greater rise in hemoglobin than could be accounted for by the amount of iron injected.

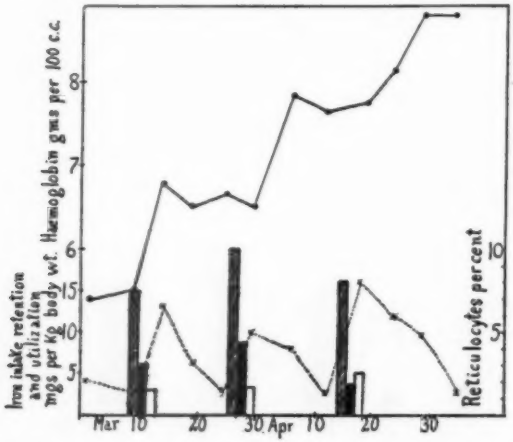
Iron intake retention and utilization mgs per Kg body wt. Hemoglobin gms per 100 c.c.
Reticulocytes per cent
Mar 10 20 30 Apr 10 20 30

CHART 10. HEMOGLOBIN AND RETICULOCYTES IN CASE OF MODERATELY SEVERE NUTRITIONAL ANEMIA (CASE 24) FOLLOWING MEDICINAL IRON

This case may be compared with case 28 (chart 9) which it closely resembled clinically, although the anemia was not so severe. As in that case, the hemoglobin rose in a series of steps, each of which followed the giving of medicinal iron. But, in contrast to what happened in case 28, in the first two periods retention greatly exceeded utilization. The fact that the duration of the rise in hemoglobin coincided with the duration of the reticulocyte response suggests that there may be some relation between the two. In the last period utilization exceeded retention, probably under the influence of copper. (See chart 5 for meaning of symbols.)

Heath, Strauss and Castle found as much as 150 per cent utilization in one of their cases, but are inclined to consider that this figure is due to the error which of necessity is large in work with iron metabolism, and which cannot be precisely determined because of the calculations which rest on assumptions regarding total circulating blood volume. Their findings, together with those of Brock and Hunter, and Heilmeyer, and the figures reported here, especially those ob-

tained under the influence of copper, introduce the possibility that one of the effects of iron is to create conditions that may lead to utilization of stored iron that had previously been apparently unavailable. Copper by itself will not do this; it is only copper in combination with iron that is effective.

Another fact, noted in the tables and graphically represented in charts 9 and 10, is that the utilization of the freshly retained iron is usually ended in about two to three weeks, even though all the iron may not have been used (chart 10) and even though the hemoglobin has not risen to its full extent, as shown by a further rise following the next "iron" period. Reimann, Fritsch and Schick (7) also record this phenomenon in one of their charts, and Brock (23) comments on the ending of the hemoglobin response after discontinuing iron, even though enough has been retained to bring the hemoglobin to 100 per cent.

Two explanations of the failure to utilize all of the retained iron appear to be possible: (1) that iron may in the course of two to three weeks be changed to a less available form and (2) that the "stimulation" caused by the medicinal iron may last only during this period. The data at our disposal seem to favor the latter of these two explanations: (1) The reticulocyte rise occurring after iron, lasts usually about two to three weeks. (2) When the amount of iron retained is too small to satisfy the demands of new hemoglobin formation stimulated by iron therapy, iron previously present in the stores is utilized, as has just been pointed out. Moreover, in the first six months of life when the conditions are such that relatively rapid hemoglobin formation takes place, iron of the "stores" is thoroughly utilized. (3) When iron therapy is continued to or beyond the end of the period of "stimulation" the rise in hemoglobin ceases in a day or two, as in Brock's case.

COMMENT

In all discussions of utilization of iron the assumption is tacitly made that the iron given therapeutically is somehow different from that already in the body—more specifically, that it is available for hemoglobin formation or for "stimulation" of red cell formation, while that already deposited in the tissues is not so available. All that we

know of iron metabolism bears this out. As has been demonstrated here after ingestion of iron, as well as by Heath, Strauss and Castle, and by Heilmeyer after injection, a small amount of medicinal iron is worth more than all the iron already in the tissues. Yet when the conditions are favorable this tissue iron becomes available. The conclusion is inescapable that one of the functions of medicinal iron is to make the conditions favorable. Whether we call this "stimulation," depends on the definition of the term. Perhaps "regulation" is a better term.

As was pointed out, the iron in the cases here studied, was first deposited in the tissues and only later appeared in the hemoglobin. We have no way of ascertaining what tissues are involved in the deposition of iron, but if one may judge from observations in animals, the major portion, at least, goes directly into the cells of the reticuloendothelial system, especially those of the liver. The amount that remains available for "stimulation" or "regulation" by direct action upon the hemopoietic tissue, must be extremely small. This renders improbable the idea that has been advanced that the "stimulating" action of iron is secondary to its utilization for hemoglobin formation within the bone marrow. Beyond this we can hardly go at present, without indulging in unprofitable speculation.

What bearing has all this on our conception of iron deficiency as applied to anemia? It has been previously pointed out that a quantitative iron deficiency has a very limited application in the explanation of anemias such as occur in rapidly growing animals, in premature babies and in others whose rate of growth is out of proportion to their birth "stores," in cases of slight reduction in hemoglobin occurring in the second half-year of life in normal babies on unsupplemented milk diets, and in cases of reduction in hemoglobin occurring somewhat earlier in babies of anemic mothers.[4] We are then forced to turn our

[4] The considerations that have led to the conclusion that there is little basis for considering the majority of cases of anemia in infancy as due to iron deficiency in a quantitative sense have no bearing on the conclusions of Heath and Patek regarding iron deficiency in adults, which may well exist on a quantitative basis, as they have shown. Of course, even in infancy if one includes cases in which the hemoglobin is only slightly reduced, say to between 8.5 and 10 gm. per 100 cc., then iron deficiency anemia in a quantitative sense certainly does exist. All

attention to the idea that iron within the body for some reason becomes unavailable, that its form changes so that it becomes more firmly incorporated in organic compounds from which it can be freed only with ever greater difficulty. The evidence in favor of this is: (1) the evident greater availability of medicinal iron, as compared with tissue iron, and (2) the fact that after two or three weeks medicinal iron is no longer utilized. But against this point of view there are equally evident facts: (1) that iron deposited at birth and shortly after birth is still available months later; (2) that after the hemoglobin level has fallen, in the course of an infection, it may rise spontaneously; and, (3) that there may be an extraordinarily rapid spontaneous rise in hemoglobin after hemorrhage or following the crises in hemolytic anemia.

Finally, then, we must consider primarily the conditions that have rendered iron unavailable. Why should it be utilized at one time and not at another? Why should it not be utilized in some cases after infection, or in cases of nutritional anemia; whereas, after hemorrhage, when the loss of iron from the body is infinitely greater, it is well utilized? These questions we may not be able to answer but they emphasize the point that deficiency or unavailability of iron is a secondary phenomenon dependent on the conditions under which the anemia has developed.

If the iron of the body is available for hemoglobin formation when the conditions are right for its utilization, and if hypochromic anemia exists, the anemia cannot be attributed only to the lack of a necessary building stone for hemoglobin formation; there must also be a lack of a substance necessary to restore the conditions essential for normal hemoglobin production. That medicinal iron is such a substance is indicated by the work reported here. But until we can show that iron is essential to this process and that no other substance

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that is claimed is that for the explanation of severe anemia in infancy quantitative iron deficiency rarely suffices, and that the mere fact that iron causes a rise in hemoglobin is not proof of a quantitative deficiency. In adults there is a time factor which is entirely lacking in babies, and as Heath and Patek have pointed out, there is also an important factor in blood loss. In babies there is only the growth factor, and this, as has been shown, is rarely sufficient to account for severe anemia.

can replace it, we have no right dogmatically to consider that the anemia in these cases is due to iron deficiency.

SUMMARY

Iron balance and utilization were studied during forty-seven short periods in infants varying in age from three weeks to twenty months, some normal, some suffering from "nutritional" anemia. During these periods, lasting five to six days, inorganic iron was given in varying but relatively small amounts. The purpose was to study the various factors that influenced retention of iron, stimulation of blood formation, utilization of iron in hemoglobin formation, and storage of iron. As a result of this work, considered together with the findings of others as reported in the literature, the following conclusions were reached.

1. Retention of iron is enhanced when the intake is above a level of 2 mg. per kg. of body weight per day. As the intake is reduced below this level the body is usually in iron balance until the intake has reached a point in the neighborhood of 0.1 mg. per kg. of body weight per day, when the balance tends to become negative. The initial hemoglobin level appears also to have some influence, in that anemic infants retain iron better than non-anemic infants. Age, within the limits of observation, had little, if any, influence.

2. When iron is taken into the body it is not eliminated for more than twenty-four to thirty-six hours after the increased intake has been discontinued. Since this is about the time required for its passage through the intestinal tract its reexcretion must be minimal. For practical purposes retention is equal to absorption at the relatively low levels of intake represented in this study.

3. The factor having the greatest influence on the reticulocyte rise is the initital hemoglobin level. The failure to demonstrate any close correlation between iron dosage and reticulocyte rise is attributed to the possible presence of other uncontrolled factors, the influence of of which on reticulocyte response may be more evident in the presence of relatively low iron intakes than when large therapeutic doses of iron are used.

4. Below six months of age, utilization depends almost entirely

upon age or rate of growth, although in some cases administration of iron causes a temporary increase in rate of hemoglobin formation.

5. Above six months of age utilization depends to a considerable extent on retention. From a consideration of figures in the literature it is felt that this correlation would probably not apply to cases in which the retention is so great that it more than satisfies the requirements for hemoglobin formation.

6. Copper causes an increase in the total utilization of iron but has much less influence on the rate of utilization. It has no influence on retention.

7. The initial hemoglobin level and the related reticulocyte rise have relatively little influence on utilization. This is, of course, at variance with the usual clinical experience and may be attributed to the use, in these experiments, of small iron doses and consequent submaximal "stimulation" so that other factors play a relatively more important rôle.

8. Iron that is taken into the body is utilized for hemoglobin formation only after the lapse of several days, *i.e.*, "storage" precedes utilization.

9. In anemic patients the percentage of retained iron that is utilized, is greater at lower levels of retention. From the figures in this work and from others in the literature it is evident that when about 2 mg. or less per kg. of body weight are retained, the utilization is about 100 per cent. Heath, Strauss and Castle found about 100 per cent utilization when iron was injected in amounts below this level.

10. The rise in hemoglobin that follows any one of these periods of iron ingestion lasts only about two to three weeks, even though all the retained iron has not been used, and the hemoglobin has not risen to its full extent. It was pointed out that the period of rising hemoglobin coincided roughly with the period of the reticulocyte increase.

The bearing that the results of this study have on the conception of iron deficiency and on the theories of the action of iron, was discussed. It was concluded that the medicinal iron had an important "regulatory" function in addition to that of supplying a building stone for hemoglobin.

CASE HISTORIES

7. Colored premature baby weighing 2200 gm. at birth. At the age of a month she had not gained any weight. This was probably due more to poor care than to any constitutional weakness, for she did very well after admission to the ward.

8. Colored premature twin weighing 2000 gm. at birth. For the first two months of life she did fairly well, gaining up to 2600 gm. Then she developed an intractable diarrhea. A short time later a hemolytic anemia appeared, becoming progressively more severe. The study of iron metabolism reported here was undertaken during the early stages of the progressive anemia. The cause of the child's nutritional disturbance and anemia, was not discovered, but was believed to be an infection of obscure nature.

9. Twin of the preceding case. She developed well and has remained normal.

10. This child, white, born at term, was admitted because he was not gaining well at home. He did well on the ward, however, and was studied for a long time to determine the effects of withholding viosterol. The periods of study reported here were carried out (1) shortly after admission and (2) after viosterol had been given.

11. This was a perfectly normal white child admitted for the purpose of study.

12. Normal white child admitted for the purpose of study.

14. Normal, slightly premature colored baby admitted for study and to avoid contact with tuberculous mother.

15. Normal white child admitted for study and to avoid contact with tuberculous mother.

16. This child was admitted at three weeks of age with convulsions, possibly due to tetany. The study of iron balance was made six weeks after recovery when she was entirely well.

17. Normal white child admitted for study and to avoid contact with tuberculous father.

21. Child admitted at about six months of age with questionable diagnosis of scurvy, the signs of which subsided spontaneously before specific treatment was given. The anemia developed under observation during a period of two months. The studies of iron balance undertaken during the period of falling hemoglobin, are contained in the preceding paper. The cause of the fall in hemoglobin is obscure: orange juice had no influence on it, and there were only occasional transient rises in temperature. Iron at the level of dosage given during the metabolic study did not influence the hemoglobin significantly, but when given in much larger doses caused prompt recovery.

22. This child was admitted to the hospital with an upper respiratory infection accompanied by a mild acute nutritional disturbance. She recovered quickly on the ward and was essentially normal at the time of metabolic study.

23. Child admitted for study because he was anemic. He had had scurvy six months previously from which he had recovered under treatment. His anemia developed following this episode. It cannot be determined from the history how

long it had been present in severe form. The red cells were 2.5 million and m.c.h. 1.6 (C. I. 0.55).[5]

24. It is not known at what age the anemia developed in this child, but from the history it is likely the onset was not before the end of the first year of life. The relatively mild and occasional infections probably played a minor part in the etiology, and the anemia, both from history and the characteristics of blood examinations, was typically nutritional in nature or, as it is coming to be called, "iron-deficiency anemia." At the time of admission the red cells were 4.2 million, the hemoglobin 5.2 gms. per cent and m.c.h. 1.25 (C. I. <0.5). She was admitted because of obstinate constipation, and she did well on the ward.

25. The child developed normally up to admission. He was admitted for pneumonia, which ran an uneventful course. His hemoglobin rose spontaneously from 6.6 to 8.5 gms. per cent in the three-week period between the termination of the pneumonia and the beginning of the metabolic study.

26. This case was one of scurvy of three weeks' duration at the time he was admitted three weeks before the first balance period was begun. Before admission he had had several nose-bleeds which might have contributed to the rather mild anemia he had on admission (hemoglobin 7.7 gm. per cent). The signs of scurvy cleared up promptly on treatment, but the anemia continued to progress, the hemoglobin falling to 5.8 gm. per cent three weeks later, and the red count from 4.8 to 3.4 million. It is possible to attribute this fall to a relatively severe respiratory infection that lasted ten days during this period.

27. This child was, as far as is known, normal up to an attack of pneumonia at about nine months, for which he was admitted. Empyema developed and the rather rapid development of anemia necessitated a transfusion. The study reported here was carried out one month later, during a normal convalescence.

28. This was a case of typical nutritional "iron-deficiency" anemia, with great reduction of hemoglobin and a mean corpuscular hemoglobin of 1.2 (C. I. = 0.43). The onset (pallor), according to the mother, was at about six months.

29. This child almost from the beginning was subject to repeated infections of more than usual severity, being admitted three times for pneumonia. During the course of the pneumonia that preceded the metabolic study his hemoglobin fell from 9.1 to 6.5 gm. per cent and showed no tendency to a spontaneous rise subsequently.

30. The child was apparently well up to the time of admission for pneumonia at seven months of age. At that time his hemoglobin was 7.0 gm. per cent. Two weeks later it was 5.8 gm. per cent. In other words, there was no tendency to spontaneous improvement in hemoglobin level during the period of observation. It is unlikely that pneumonia was the only factor in the low hemoglobin.

31. The child was admitted for pneumonia at nine months. During the first three days in the hospital, the hemoglobin fell from 8.1 to 6.7 gm. per cent

[5] m.c.h. = mean corpuscular hemoglobin, expressed in micromicrograms.

and there was no tendency to spontaneous rise in three weeks of preliminary observation.

32. The metabolism study in this child followed recovery from a severe nutritional disturbance.

33. This was a normal child admitted for the study of iron metabolism.

34. This child was admitted for a mild upper respiratory infection with diarrhea. At the time of the study of iron metabolism she was virtually normal.

35. A case of osteogenesis imperfecta, with multiple skin abscesses that were accompanied by considerable anemia. The abscesses, however, had cleared up, and the hemoglobin had returned to normal at the time of the study. A tendency to negative iron balance was noticeable in the preliminary period of study.

36. Normal child admitted for study.

37. This child was admitted with pneumonia which ran an uneventful course. The hemoglobin which was 6.8 gm. per cent on admission rose spontaneously after the termination of the infection. The reduction of red cells to 2.8 million makes it appear likely that the anemia in this case was due to the infection.

38. This was a case of congenital hemolytic jaundice with increased urobilin excretion in the stools. He was studied during a more or less quiescent period.

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THE RELATION OF SYPHILIS TO NEPHRITIS*

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The occurrence of nephritis in a patient with syphilis has always offered to physicians a confusing problem in diagnosis and one with consequences of great practical importance depending upon its correct solution. If the nephritis is viewed as a manifestation of the syphilitic infection then vigorous antisyphilitic treatment should be pushed without delay; if, on the other hand, the nephritis is regarded as having no direct relation to the syphilitic infection, but as a chance occurrence of nephritis in a syphilitic, then great caution should be exercised in administering even the most conservative treatment for the syphilis. The problem is not at all a new one. Nearly one hundred years ago Rayer (1) confessed to being greatly puzzled by it. He reports the case of a patient with syphilis and albuminuria and after a lengthy discussion admits that he is unable to reach a positive conclusion as to the exact relation the one bore to the other. The uncertainty persists to the present day. One group of physicians consider syphilitic nephritis to be a clear-cut clinical condition and under appropriate circumstances does not hesitate to make the certain diagnosis. Another group doubts that syphilis is ever the direct cause of nephritis and is unwilling to allow that the diagnosis can ever be made with confidence.

It has become customary to divide nephritis into three main groups, namely, glomerular nephritis, vascular nephritis, and lipoid nephrosis. There are clinical and pathological features which in most instances permit us to distinguish precisely between them. These features are well recognized and I shall not stop to review them. The question which now holds our interest is to decide whether or not syphilis can ever be the sole cause of these types of nephritis. This question would be promptly and satisfactorily answered were it sufficient to submit it

* This paper is based in large part upon a paper read before the Section on Medicine, Southern Medical Association, November 21, 1935.

to the test of demonstrating the presence or absence of spirochetes in the renal lesions. Unfortunately this test is altogether unreliable. No method of staining tissues has been devised which will uniformly demonstrate spirochetes when they are present. Indeed only rarely are pathologists able to demonstrate them in undoubted syphilitic lesions. More than this, it is possible that spirochetes may be present in the kidneys of syphilitic patients without producing nephritis (2). Therefore even though spirochetes were found in the kidneys of a syphilitic patient with nephritis this might not definitely prove that the nephritis was caused by the syphilis. The pathological features of syphilitic lesions have characteristic marks by which they may be recognized but these marks are not absolutely distinctive even though their presence makes it very probable that the lesion is syphilitic. Unfortunately the reaction to other infectious agents may produce similar lesions and in many instances an absolute distinction is impossible from a study of the gross and microscopical features alone. For instance, I need merely mention the difference of opinion which still exists amongst pathologists regarding the syphilitic nature of certain myocardial lesions. Finally, it is possible that certain renal lesions may be due to syphilis even though they may show no pathological changes in any way characteristic of syphilis. It is well known that the kidneys may react very violently to certain toxic products of infections although there is nothing specific about the appearance of the renal damage to tell us precisely what organism was the infectious agent. For instance, a patient has streptococcus tonsillitis and presently develops glomerular nephritis. From this sequence of clinical events we conclude that the nephritis is a reaction to the streptococcus infection although there is nothing in the appearance, investigation of the kidneys or in the bacteriologic investigation which independently would establish this diagnosis. Similarly the diagnosis of syphilitic nephritis is very difficult to establish. It may never be established with absolute certainty. All we can anticipate is that the diagnosis may at times be made with a high degree of probability.

In reviewing the medical literature on the subject, one finds lengthy discussions seeking to demonstrate that syphilis may attack arterioles, glomeruli and tubules in such a way as to reproduce clinically the

three common types of nephritis. On the other hand, there are equally lengthy discussions which emphasize the contradictory nature of pathological evidence and its unreliability. Lamierre and Vallery-Radot (3) review the disputed points in great detail and conclude that pathologically the kidneys from suspected cases of syphilitic nephritis show no specific lesions.

HEMORRHAGIC AND VASCULAR NEPHRITIS

There is little or no evidence that syphilis can produce the specific picture of ordinary hemorrhagic nephritis. It is true that hemorrhagic nephritis often occurs during the florid stages of secondary or congenital syphilis, but, as Rich (4) has pointed out, the kidneys from these cases are pathologically like any non-syphilitic hemorrhagic nephritis and the probable explanation lies in the fact that the syphilitic lesions (*e.g.*, ulceration of the nasal and pharyngeal mucous membranes) serve as portals of entry for some bacterial infection now known frequently to precede the development of hemorrhagic nephritis. If this explanation be correct then the beneficial effect of anti-syphilitic treatment reported in these cases should be attributed to the healing of portals of entry for the causative infection.

As regards vascular nephritis there is even less evidence that syphilis plays an etiological rôle. Localized syphilis of the renal arterioles has never been proved pathologically though there is some evidence that it may occur. Syphilis has often been suggested as one cause of hypertension and widespread arteriosclerosis of which arteriosclerotic nephritis is a part. Reliable statistical evidence has never substantiated this view.

LIPOID NEPHROSIS

There is more persuasive evidence that syphilis may play an etiological rôle in lipoid nephrosis. Herrmann and Marr (5) have recently published a comprehensive review of the subject. Their material consisted of 1040 syphilitic patients, 7.1 per cent of whom had significant albuminuria. None of thirty-one patients with primary syphilis had any urinary abnormality. Of the 74 cases of albuminuria with either secondary or tertiary syphilis there were four with massive albuminuria and edema. The first case was a seventeen year old boy

who developed the picture of lipoid nephrosis four weeks after the appearance of genital lesions. After three doses of sodium bismuth tartrate the edema and albuminuria disappeared, and with continued antisyphilitic treatment they did not return. The second and third cases were examples of nephrosis and late syphilis and although they improved under antisyphilitic treatment the albuminuria never completely disappeared. The fourth patient received no antisyphilitic treatment. Wosika and Thurmon (6) have reported three cases with late syphilis and nephrosis in which the renal disease cleared up after prolonged antisyphilitic treatment. Moore (7) has recorded an extraordinarily severe nephrosis in a young woman with secondary syphilis which disappeared entirely after the administration of neoarsphenamine. There are other similar cases recorded in the review of Herrmann and Marr (5).

From these clinical observations it is clear that certain syphilitic individuals with the nephrotic type of Bright's disease improve or lose their albuminuria after antisyphilitic treatment. However, as previously stated, the pathological changes in the kidneys of similar untreated cases, do not serve to distinguish them from the usual form of non-syphilitic lipoid nephrosis.

TRUE SYPHILITIC NEPHRITIS

From the pathological point of view Rich (4) has recently recorded the finding of interesting kidney lesions in syphilitic subjects. These lesions were found in the kidneys in 13 (6.5 per cent) of 200 autopsies at which clearly recognized syphilitic lesions (chiefly aortitis) were found in other organs. In 400 control autopsies at which no syphilitic lesions were found in other organs, the kidney lesions under discussion were found twice. During life both of these patients had had positive Wassermann reactions. Four additional cases brought the total to 19 and in all there was definite proof that the patients had syphilitic infections. Since this report, the same peculiar lesion has been repeatedly encountered in autopsies on syphilitics in this hospital.

All of the lesions described by Rich are similar in nature though they vary considerably in extent. In mildly affected kidneys the lesions may be seen as greyish-yellow flecks dotting the cortex of the kidney. In more extensively involved kidneys the surfaces are

roughened and the striae are distorted. Microscopically the lesions consist of dense accumulations of mononuclear cells in the renal interstitial tissue. There are also perivascular accumulations of lymphocytes with occasional plasma cells and macrophages. These accumulations of cells assume a characteristic spherical arrangement and in many places compress the tubules and sometimes protrude into their lumina. A second characteristic of the nephritis is the occurrence of lipoid laden macrophages and undoubted cholesterine crystals in tubules adjacent to the lesions. As Rich points out the final proof of the syphilitic nature of these peculiar lesions must await the demonstration of spirochetes which have not as yet been found. But the lesions are like other known syphilitic lesions and the fact that they are found only in patients with syphilis leaves little doubt that they are specific.

Whereas the observations of Rich almost certainly establish the fact that syphilis may specifically attack the kidney, they do not clarify the uncertainty regarding the occurrence of clinical syphilitic nephritis. The lesions which Rich has described would have to be very widespread to effect any significant alteration in kidney function. In four of his cases the pathological changes in the kidneys were very widespread but during life congestive heart failure had clouded the picture, and the patients had not been thoroughly studied from the standpoint of the kidney condition. However in a more carefully studied case, reported by Bauer (8), the peculiar nephritis observed by Rich was found. The patient was a 41-year-old male who had acquired syphilis 16 years before. He had hypertension, moderate albuminuria, hyaline cylindruria, a positive Wassermann test and a rapidly rising non-protein nitrogen of the blood. He died in uremia. The changes in the kidney were widespread and quite similar to those recorded by Rich. There was no other clinical or pathological explanation for the rapidly developing renal insufficiency.

CLINICAL DIAGNOSIS

It is reasonable to conclude from a study of the literature that many of the reported cases of syphilitic nephritis were erroneously diagnosed. It is certain that cases with nephrosis and syphilis, whether or not the two conditions are related etiologically, reveal at autopsy

lesions which do not differ from those of non-syphilitic nephrosis. Even if spirochaetes are demonstrated in the tubular lesions characteristic of nephrosis, as in the case of glomerular nephritis reported by Miller and Hays (9), the syphilitic etiology of the lesions will still be unestablished. Since it is generally agreed that if syphilis produces nephritis it produces the nephrotic type, the final proof of the argument must await experimental confirmation. It must be demonstrated that nephrotic lesions occur in the kidneys of experimental animals as a result of infection with the *spirochaeta pallida*.

Nevertheless occasionally during the florid, acute or even late stages of syphilitic infections the clinical features of nephrosis appear. Often in these cases, following the administration of antisyphilitic drugs, the signs and symptoms of nephrosis are greatly ameliorated and in a number of cases recovery occurs. This is certainly not the usual course of nephrosis in non-syphilitic adults. Therefore it seems warranted to conclude that syphilis can initiate the onset of nephrosis. There are two possible explanations for the association of these two diseases in the same patient. The first is that the causes of nephrosis are multiple and syphilis is one of them, and the second that the syphilitic infection serves to admit an unknown agent capable of producing lipoid nephrosis.

Those instances of nephritis secondary to amyloid deposits in the kidneys are in a somewhat different category. Any widespread chronic infection may lead to amyloidosis with manifestations of nephritis but the variety secondary to syphilitic infection has no distinguishing features.

REPORT OF CASES

In consideration of the facts discussed above it is apparent that when the possibility of syphilitic nephritis presents itself the diagnosis cannot be made without the most critical scrutiny. For this reason it does not seem out of place to present the clinical records of two patients, one of whom had albuminuria and cylindruria with early syphilis, and the other a similar abnormality in association with late syphilis. There are certain peculiar relationships which exist in patients in whom syphilis and nephritis are combined, whether or not the relationship is a causal one. It seems justifiable to conclude from

the foregoing discussion that the matter is not yet settled, and I believe the following cases serve to emphasize further the present state of uncertainty.

Case 1. (Unit History 27,694) R. W., a colored male laborer, 25 years of age, was admitted to the Johns Hopkins Hospital October 28, 1929 and was discharged November 18, 1929. His chief complaints were swelling of the ankles, breathlessness and a skin rash. The family history contributed no data of importance. In the past the patient had enjoyed unusually good health. Indeed, his only illness was an attack of acute gonorrhea in 1924 for which he was examined and treated in the Johns Hopkins Hospital Dispensary. At the time of that examination the urine contained many pus cells but neither albumen, blood nor casts. The urethritis cleared up rapidly as a result of appropriate local treatment and the patient had no other genito-urinary symptoms until the onset of the condition for which he was admitted to the hospital.

Present Illness: In the early part of August 1928 the patient developed a sore on the penis. The sore was treated with medicine procured from a pharmacist and it healed in about five days. Usual good health then continued until three weeks before admission to the hospital when the patient contracted an ordinary head cold which was followed by a "little hoarseness" and cough. There was no sore throat. Two weeks before admission the patient developed a papular skin rash which spread over the entire body during the course of the next week. The patient denied feeling badly at the time the rash appeared, and he continued to work at the arduous task of a laborer in a steel mill. At about the time the skin rash appeared nycturia set in. No change was detected in the amount of urine voided nor did it ever appear to contain blood. One week before admission the feet and ankles began to swell. The swelling rapidly spread upwards and involved the entire legs, and the external genitalia. At first the swelling would subside over night but for the three days before admission it increased rapidly. The patient became quite weak and noticed breathlessness on exertion. There was never puffiness of the face or eyes and never nausea, vomiting, headache or visual disturbances.

Physical Examination: Temperature, 102.2°; pulse, 64; respirations, 22; blood pressure, 155/90; weight 79.25 Kg.

General appearance: The patient was a young colored man who lay partly propped up in bed in no apparent distress. There was no cough. He was mentally clear and cooperative.

Skeletal system: There were no deformities, clubbing of the fingers or abnormalities in the joints.

Skin: The mucus membranes were of good color and there was no cyanosis. There was no puffiness of the face or eyes and no apparent edema of the upper extremities. There was very slight pitting edema over the sternum and massive edema of the lower abdominal wall, the lower back, the external genitalia and the

lower extremities. With the exception of the face, the palms of the hands and the soles of the feet, the skin of the entire body was the site of a purplish, papular rash. Some of the papules showed central exfoliation but no bleeding or exudation.

Lymphnodes: All the superficial lymphnodes, while not greatly enlarged or tender, were readily palpable.

Eyes: Normally prominent. No periorbital swelling. The pupils were equal and reacted readily to light and accommodation. The extra-ocular movements were normal.

Ocular fundi: The optic discs were sharply outlined and of normal color. The arteries and veins appeared normal. There were no exudates or hemorrhages and the retinae were not edematous.

Mouth: The tongue was slightly coated but otherwise normal. The teeth showed evidence of moderate caries. The tonsils were not enlarged and were not inflamed except over the surface of the left tonsil where there was a very superficial gray ulcer 0.5 cm. in diameter.

Neck: The thyroid gland was normal. The trachea was in the midline and there was no tracheal tug. The veins were not engorged.

Chest: Well formed and symmetrical. The percussion note was everywhere resonant and the breath sounds were of normal intensity and clear. There were no rales.

Heart: The cardiac impulse was quite forceful and was readily seen in the fifth interspace, 14 cm. from the mid-sternal line. In the fourth space dulness extended 12 cm. to the left and 3 cm. to the right of the midline. At the apex the first sound was loud and was followed by a short systolic murmur which could not be heard over the rest of the heart. No other murmurs were heard. The aortic and pulmonic second sounds were normal. The cardiac rhythm was regular and the radial arteries felt normal.

Abdomen: The abdomen was distended and there seemed to be a small peritoneal effusion. The liver, spleen and kidneys could not be felt.

Genitalia: Normal except for massive edema.

Neurological: The cranial nerves functioned normally. There was no muscle wasting or weakness and no sensory disturbance. The deep reflexes were equal and active.

Laboratory Examinations. Blood count: Hemoglobin 85 per cent; red blood cells 4,600,000; white blood cells 8,000. Normal smear and differential formula. *Urine:* Amber, specific gravity 1.016; sugar negative; albumen 4+; Microscopic: Rare red blood cells; moderate number of white blood cells; few hyaline and granular casts. Upon many subsequent examinations red blood cells were not observed. *Blood Wassermann:* Strongly positive in both ice-box and water-bath fixations. *Blood chemistry:* Nonprotein nitrogen 34 mgm. per 100 cc.; creatinine 1.7 mgm. per 100 cc.; uric acid 4.1 mgm. per 100 cc.; NaCl 526 mgm. per 100 cc.; total plasma protein 5.56 gm. per 100 cc.; A/G ratio 30/70.

Phthalein test:

First hour	55 per cent
Second hour	15 per cent
Total	70 per cent

Urea clearance test:

Urine 1. 49 per cent of normal maximal clearance.

Urine 2. 46 per cent of normal maximal clearance.

Electrocardiogram: Rate 58: Rhythm sinoauricular; P-R interval 0.17 seconds. QRS interval normal. Normal sinus rhythm. T1 and T2 upright. T3 inverted. Ventricular complexes sharply defined.

Throat culture: Mixed flora. No haemolytic streptococci.

Dark field examination of urine: Repeatedly negative for spirochaetes. No doubly refractile bodies seen.

Two hour concentration test:

Range of specific gravity 1.008–1.020.

Day:Night ratio—1.095:740

Course in the hospital: Upon admission to the hospital the patient was put to bed at absolute rest. The fluid intake was limited to two liters daily and the diet consisted of fruit juices, mild and cooked cereals without free salt. The patient was never sufficiently distressed to require sedative drugs. Improvement in the patient's condition began almost immediately and by the sixth day he had lost 12.75 kilograms of body weight as a result of a copious diuresis. On the sixth day of admission an initial injection of 0.1 gm. Neoarsphenamine was given intravenously. This dose was repeated on the eighth day and was increased to 0.2 gm. on the eleventh day and again on the seventeenth day. The edema and albuminuria disappeared with amazing rapidity. Four days after admission the urine contained no albumen and there was no demonstrable pitting edema. The left border of cardiac dulness had by this time moved inward 3 cm. and two days later a teleoroentgenogram showed a total heart width of only 11.6 cm. Twenty-one days after admission the skin lesions had entirely disappeared, as had the albuminuria and edema. The patient felt well and he was allowed to leave the hospital.

After leaving the hospital the patient returned regularly to the Out-patient Department for antisyphilitic treatment and at no time did he show any edema or any abnormality of the urine.

Diagnosis: The patient obviously had very active secondary syphilis and, associated with this, some disease either of his kidneys or circulatory apparatus leading to the development of massive edema, albuminuria and cylindruria. Most of those who saw the patient concluded that he did not have ordinary hemorrhagic nephritis but rather some form of acute nephritis associated with syphilis. Others felt that the edema and albuminuria were circulatory in origin. Certain considerations are in favor of the latter explanation. The edema was confined to dependent parts of the body. The heart was greatly dilated on admission to the

hospital and very rapidly returned to normal size as the patient improved. The degree of cylindruria was not proportional to the albuminuria and there were no cellular casts, both findings uncommon in an acute nephritis. Finally the albuminuria and edema both disappeared completely after four days rest in bed with limitation of fluid and salt intake, before the administration of any antisyphilitic drug. It seems unlikely that a nephritis sufficiently diffuse to produce such extensive protein loss and retention of water could heal within four days. On the other hand rapid improvement in circulatory efficiency is not uncommon in cases of congestive heart failure.[1] Nevertheless, it may be of significance that this patient did not have the enlarged liver of chronic passive congestion, and that the degree of albuminuria was more than one usually sees even in severe grades of congestive heart failure. Whitehill, Longcope and Williams (10) have recently emphasized the frequent occurrence of transitory cardiac enlargement, and signs of myocardial insufficiency in cases of acute hemorrhagic nephritis. It is not inconceivable, therefore, that the cardiac changes in the present case were secondary to, or associated with an acute renal disturbance, possibly of syphilitic origin.

Case 2. (Unit History 28,428) R. A., a colored woman, 27 years of age, was admitted to the Johns Hopkins Hospital December 10, 1929 and discharged February 22, 1930. She complained of failing vision, headache and pain in the chest. The family history contributed nothing of importance. The patient's mentality was greatly retarded and the accuracy of her history was always strongly questioned. Nevertheless, she stated that she had regularly enjoyed reasonably good health. All the patient's life she had suffered from two or three attacks of severe sore throat each winter. Quite frequently the attacks of sore throat were followed by bronchitis forcing her to bed for a week or two but she apparently always recovered her usual health. Prior to the onset of the present illness the patient had never had any urinary symptoms and, to her knowledge, she had never had any venereal disease. The patient had been pregnant seven times and had miscarried three times before the fifth month. Her last pregnancy resulted in the birth of a healthy boy at term in 1926.

The present illness began rather vaguely in August 1929, four months before admission to the hospital. Weakness and breathlessness on exertion interfered with her accustomed activity. The patient noticed an increasing thirst and with it the necessity to void three or four times each night. Early in September 1929, after a large meal, she had a "bad pain over the heart" which persisted for two days. She had with this attack a very severe headache which caused her to vomit frequently. From this attack until the time of admission to the hospital the patient was apparently confined to bed almost constantly. It was impossible to be

[1] If congestive heart failure be the proper explanation of the patient's illness, it may have been due to extensive disease of the myocardium caused by the florid syphilitic infection.

certain of details but headache, nausea, vomiting, weakness, failing vision and a loss of 30 pounds in weight were the outstanding symptoms.

Physical Examination: Temperature, 99°; pulse, 110; respirations, 20; blood pressure, 155/90; weight, 116 lbs.

The patient was a well developed negress lying flat in bed in no respiratory distress. She had obviously lost a great deal of weight. Though she complained of severe headache she laughed and giggled in a silly way as if she were not greatly distressed.

Skin: There were a few small areas of leukoderma but no skin rash. The mucous membranes were very pale. There were no mucous patches. There was no pitting edema.

Skeletal system: There were no gross deformities of any of the long bones or joints. The tibiae and the clavicles were neither roughened nor tender.

Eyes: Prominent but symmetrically so and with some periorbital swelling. The pupillary reactions were normal as were the extra-ocular movements.

Ocular fundi: The media were clear but the disc outlines were greatly blurred both by retinal edema and very definite elevation of the nerve heads. There were numerous small hemorrhages both fresh and old about both discs but the peripheral fundi were clear. The retinal veins were abnormally full. Dr. William H. Wilmer, who saw the patient in consultation, considered the retinal arterioles normal.

Nose: Adequate breathing space. No ulceration of the septum. No discharge.

Mouth: No lesions on the lips, tongue or mucous membranes. Moderate dental caries. The tonsils were small and scarred but not inflamed.

Lymphnodes: There were a few small palpable nodes at the angles of the jaws but no generalized enlargement.

Neck: Trachea in the midline. No tug. No engorgement of the veins. Thyroid gland slightly larger than the average size but not nodular.

Chest: Well formed and symmetrical; free movements; normal descent of the lung bases. Lungs clear to percussion and auscultation.

Heart: No prominence of the precordium. No abnormal pulsation, shock, or heave. No thrill. The cardiac dulness extended to the left 10 cm. from the midsternal line in the 5th interspace; to the right 3 cm. in the 4th interspace. There was a soft apical systolic murmur but no other murmur. The aortic second sound was quite loud but not bell-like in quality. The cardiac rhythm was regular and the peripheral vessels did not feel abnormal.

Abdomen: Walls soft and flabby. No tenderness or muscle spasm. The liver, spleen and kidneys were not felt. No free fluid.

Pelvis: Normal findings.

Nervous system: Station and gait normal. No ataxia, normal cranial nerves except as recorded above regarding the optic nerves. No sensory or motor disturbances. Normal tendon reflexes. No evidence of meningeal irritation.

Laboratory Examinations. Blood count: Hemoglobin 50 per cent; Red blood

cells 2,870,000; white blood cells 9,550. The differential formula was normal but the red blood cells varied considerably in size and were pale. There were no evidences in the blood smear of regeneration. *Urine:* Pale straw in color. Specific gravity 1.010; sugar negative; albumen trace. Microscopic: Few white blood cells. No red blood cells. Very occasional thin hyaline cast. *Blood Wassermann:* Strongly positive in both water bath and ice box fixation. *Blood chemistry:* Nonprotein nitrogen 50 mgm. per 100 cc.; creatinine 2.6 mgm. per 100 cc.; sugar 95.0 mgm. per 100 cc.; uric acid 4.4 mgm. per 100 cc. CO_2 combining power 34.3 vols. per cent. Refractive index 1.3518.

X-rays: Sinuses clear. Chest: Lungs clear; heart and aorta within normal limits. Transverse diameter 26 cm. Skull: Negative.

Phthalein test: (Dye given intravenously).

Excretion first hour—500 cc.	5 per cent
Excretion second hour—140 cc.	10 per cent
Total	15 per cent

Urine culture: No growth. *Throat culture:* Heavy growth of streptococcus viridans but no hemolytic streptococci of Beta type.

Urea Clearance Test:

Urine #1	18 per cent of normal standard clearance
Urine #2	17 per cent of normal standard clearance

Two Hour Concentration Test:

Range of specific gravity	1.005–1.010
Day:night ratio	930:800

Basal Metabolic Rate: −6 per cent

Course in the Hospital: Throughout the period of observation the temperature, pulse and respiratory rates remained within normal limits. The patient complained a great deal of headache and was frequently nauseated. The evidences of optic neuritis were even more striking a week after admission to the hospital. On December 18, 1929 a lumbar puncture was performed. The fluid seemed to be under increased pressure though this was not accurately measured. It was colorless but contained 139 mononuclear cells per cu. mm. The Wassermann reaction on the fluid was negative in all dilutions and the colloidal mastic test gave no precipitation in dilutions of 1:4 through 1:2048.

For the first ten days in the hospital treatment consisted of a general diet the protein content of which was reduced to 60 Gm. daily. The diet was not salt-free and the patient was allowed to add salt to her food. Fluids were forced so that the average daily intake was three liters. Mild sedatives and analgesics were administered to control the headache. The patient's condition changed very little. Indeed she became more listless and drowsy.

After the spinal fluid findings became known intensive antisyphilitic treatment was begun in the form of potassium iodide by mouth and neoarsphenamine by vein, the latter first in small doses and finally in doses of 0.45 grams. This treatment was followed almost immediately by remarkable improvement. The

headache, nausea and vomiting rapidly ceased. The appetite returned. The blood pressure and the blood non-protein nitrogen fell. The blood CO_2 combining power rose. The excretion of phthalein, and other tests of renal function improved rapidly. The spinal fluid was examined on three separate occasions. There was always an increase in the number of mononuclear cells but not as great as on admission. The Wassermann reaction on the spinal fluid was persistently negative.

At the time the patient left the hospital on February 22, 1930, she was subjectively well. The papilledema had almost subsided and she noticed no disturbance of vision. The blood pressure was normal. The body weight had risen from 116 pounds on admission to 126 pounds. The urine was concentrated and was free from albumen and casts. The anemia had improved remarkably and the tests of renal function had almost reached normal levels.

Course after discharge: The patient was instructed to return to the Out-patient Department regularly for antisyphilitic treatment. This she did only for a short time. She received one course of six weekly injections of 0.45 grams of neoarsphenamine and four injections of 0.2 grams of bismuth. She then stopped treatment and the most searching efforts failed to discover her whereabouts. Finally a year later, in May 1931, she was found. The patient stated that she had been in perfect health and doing all the work necessary to care for a large family. She had had no headache, nausea or vomiting. Her vision had remained normal. She had had no dyspnea, palpitation or edema. There had been no urinary symptoms. Her appetite and digestion had been excellent and she had gained 30 lbs. since leaving the hospital.

Physical Examination: Temperature 98°; pulse 65; respirations 15; blood pressure 130/75; weight 151 lbs. *General appearance:* The patient looked quite healthy. There was no edema and there were no skin lesions. *Eyes:* They were normally prominent. The pupils were equal and regular and they reacted actively to light and accommodation. The extraocular movements were normal. *Ocular fundi:* The media were clear. There was no general retinal edema. There were no hemorrhages or exudates and even the finest macular arterioles appeared thin and delicate. The discs were hazy in outline but they were of normal color. *Mouth:* Nothing remarkable. *Lymph nodes:* Not enlarged. *Chest:* Well formed. Free movements of both lung bases. The lungs were clear to percussion and auscultation. *Heart:* There was no abnormal pulsation. Dulness extended 8.5 cm. to the left in the fifth interspace. The retrosternal dulness was not increased. The heart sounds were everywhere of normal quality. There were no murmurs. There was nothing remarkable about the pulses or the vessel walls. *Abdomen:* Nothing abnormal. *Nervous system:* The station and gait were normal. The cranial nerves all functioned normally. The deep reflexes were all equal and there were no abnormal reflexes. There was no ataxia and no sensory disturbance.

Laboratory Examinations. Urine: Clear; insufficient quantity to measure specific gravity. Albumen negative; sugar negative. Microscopic: No casts or red blood cells; only a moderate number of leukocytes.

Blood: Hemoglobin 85 per cent; Red blood cells 4,700,000; white blood cells 6,200. The smear and differential formula were normal.

Blood Wassermann: Strongly positive.

Phthalein test:

First hour excretion	50 per cent
Second hour excretion	15 per cent
Total	65 per cent

Blood chemistry: Nonprotein nitrogen 30 mgm. per 100 cc.; creatinine 1.7 mgm. per 100 cc.; uric acid 2.6 mgm. per 100 cc. NaCl 500.0 mgm. per 100 cc.

Diagnosis: This case is even more puzzling than the first. Obviously the patient had late syphilis. Also there were all the manifestations of grave renal insufficiency associated with hypertension, minimal albuminuria and cylindruria and no disturbance in water excretion. Clearly, circulatory failure cannot explain the picture. Unfortunately one possible explanation was not considered until too late for certain essential studies to be made. The headache, nausea, vomiting, optic neuritis and spinal fluid findings, in spite of the negative spinal fluid serology, appear to have been due to syphilitic meningitis. That such vomiting as the patient had may lead to loss of electrolytes with acidosis and grave renal insufficiency is well known (Blum *et al.* (11) and Landis *et al.* (12)). This possibility was not thought of until antisyphilitic treatment had been started, the vomiting had ceased and the manifestations of renal insufficiency had begun to lessen. When the sodium chloride of the blood was finally determined the figure was within normal limits.

It was concluded by many who saw this patient that she suffered from some form of syphilis of the kidney. The urinary findings were never such as to suggest an inflammatory lesion of the glomeruli or the tubular changes of lipoid nephrosis. The abnormalities in the urine were much like those found in the end stages of arteriosclerotic nephritis. It was suggested that in this patient there was widespread scarring of the kidney from syphilis of the renal arteries. This explanation is quite untenable in view of the complete and permanent return of renal function.

Finally, the possibility remains that this was an example of the specific syphilitic nephritis described by Rich. At the time this patient was ill Rich had not published his observations and the condition was unknown. No peculiar crystals were observed in the urine which might have been the cholesterin crystals so characteristic of the nephritis Rich described. If the lesions of Rich had been sufficiently widespread to produce such advanced renal insufficiency it does not seem logical that complete restoration of normal kidney function could have been brought about by antisyphilitic treatment.

CONCLUSIONS

Though the possible existence of syphilitic nephritis has been considered for many years only very recently has this possibility been established (Rich (5) and Bauer (8)). This is a peculiar form of nephritis and certainly only rarely (Bauer) is it sufficiently widespread to produce the clinical symptoms of nephritis. It does not explain the many cases of severe nephritis which occur in syphilitic patients who are strikingly benefitted by anti-syphilitic treatment. The majority of these cases are instances of nephrosis and as yet there is no definite pathological proof that syphilis can specifically attack the kidney and produce the lesions characteristic of nephrosis.

For these reasons a clinical diagnosis of syphilitic nephritis should be made only after the most searching study for some other explanation. The two cases presented emphasize this point clearly.

These conclusions are not intended to decry the use of antisyphilitic treatment in patients who have combined syphilis and nephritis. The two cases presented argue strongly to the contrary. It is well known that the hemorrhagic nephritis of congenital syphilis may be greatly benefitted by antisyphilitic treatment, presumably by closing portals of entry for other infections. Furthermore many cases of nephrosis and syphilis are greatly benefitted, and some are even cured by antisyphilitic treatment. Whatever may be the mechanism by which the renal lesions are produced, for all practical purposes, some of these patients may be regarded as having syphilitic nephrosis. Finally, as Rich has pointed out the lesions which he has described are by no means uncommon. In patients with syphilis and some form of non-syphilitic nephritis there may be superimposed this peculiar form of specific nephritis. If allowed to advance it will undoubtedly hasten the advent of renal insufficiency in kidneys already damaged by some other process.

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THE INACTIVATION OF TESTOSTERONE PROPIONATE AND ESTRONE IN RATS[1]

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Previous observations have indicated that the semi-synthetic and naturally occurring estrogens are rendered inactive by the liver. Zondek (1) noted that estrogen was inactivated when it was incubated *in vitro* with liver. Israel, Meranze, and Johnston (2) showed that ketohydroxy estratriene was inactivated in a heart-lung-liver perfusion system, but not in a heart-lung perfusion system. Golden and Sevringhaus (3), using normal female rats, found that if the ovaries were transplanted to the mesentery the animals gave no evidence of coming into heat. Estrus reappeared in these animals when the ovaries were retransplanted to the axilla. There have been no similar observations concerning the androgens.

The pellet technic of administration (4) affords a method for determining the effect of the liver, spleen, and other organs upon the activity of testosterone propionate and estrone. This method permits the introduction of these substances in various sites in the body, as small compressed pellets.

In the present study, these pure crystalline substances were compressed into small pellets, 1.83 mm. in diameter, and varying in weight from 1 to 6 mg. Groups of male and female rats, varying in age from 90 to 150 days, were castrated. Male castrates were used only after 2 or 3 weeks had elapsed to permit atrophy of the genital system. Female castrates were not used until the vaginal smears showed permanent anestrous.

In each of the groups of castrated male rats a weighed testosterone

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[2] National Research Council Fellow in Pathology.

propionate[3] pellet was inserted into the spleen, into the subcutaneous tissues of the back, or into the liver. After an interval of 3 weeks, the effect of the pellet was determined by weighing the seminal vesicles, prostate, penis, bulbo-urethral muscles, and preputial glands of each animal immediately after autopsy, and comparing the weights with those observed in untreated control animals. The average daily absorption of the androgen was ascertained by recovering each pellet, and determining its weight after it had been dried and cleaned of adherent tissue.

Three groups of castrated male rats were used to study the effect of the liver and spleen on the specific action of testosterone propionate. To remove the spleen from the portal circulation it was transplanted between the skin and abdominal muscles (5). The date of transplantation of the spleen, implantation of the pellet, ligation of the pedicle containing the splenic artery and vein, and termination of each experiment is tabulated, for each of these three groups of animals, in Table I. This same information is listed on the basis of elapsed time in Table II.

The pellet in the normally situated spleen is unable to exert its specific effect upon the organs of the genital tract, as indicated in Table II. The weights of the genital organs of this group are similar to the weights of the corresponding organs of the castrate control group. The weights of the organs of both these groups are significantly less than those of the corresponding organs of the group with the pellet situated in the subcutaneous tissues of the back. In Groups A and B, the weights of the genital organs are similar, and approximately two-thirds of the corresponding weights in the group with subcutaneously implanted pellets.

In Group C, where peri-splenic adhesions were given more time to form because the spleen was transplanted 2 weeks before the pellet was implanted, and where ligation of the pedicle took place 2 weeks before the animals were sacrificed, the weights of the genital organs are approximately the same as the corresponding weights in the group with subcutaneously implanted pellets. The difference in weight between the respective organs in Groups A and B, and Group C, may

[3] Supplied through the courtesy of Dr. Robert C. Mautner of Ciba Pharmaceutical Products, Inc.

be accounted for by the longer duration of stimulation by the testosterone propionate which was absorbed after the pedicle had been ligated in the animals of Group C. These results indicate that testosterone propionate implanted in the spleen is inactive unless the spleen is transplanted and its pedicle ligated.

TABLE I

Dates of operative procedures

Testosterone Propionate Pellets Implanted in Spleen

	TRANSPLANTATION OF SPLEEN	IMPLANTATION OF PELLET	LIGATION OF SPLENIC PEDICLE	ANIMALS SACRIFICED
Group A	4-19-39	4- 5-39	5-3-39	5-10-39
Group B	4-19-39	4-19-39	5-3-39	5-10-39
Group C	4-15-39	4-29-39	5-6-39	5-20-39

TABLE II

Inactivation of testosterone propionate in castrated male rats

SITE OF IMPLANTATION	ANIMALS		SPLEEN		PELLET		AVERAGE WEIGHT OF ORGANS IN MG.				
	No.	Ave. wt.	Period of transplantation	Period of ligation	Period of implantation	Ave. daily absorption	Seminal vesicles	Bulbo-urethral muscle	Prostate	Penis	Preputial glands
		grams	*wks.*	*wks.*	*wks.*	*mg.*					
None (castrate controls)	4	175					22.9	149.7	52.2	84.9	49.4
Spleen in normal position	7	246			3	.064	29	124	71	118	48
Group A	4	247	3	1	5	.058	195	435.8	303	202	77.7
Group B	3	168	3	1	3	.070	188	428.5	308	155.9	74.1
Group C	3	188	5	2	3	.073	341.6	539	515	198.6	86.6
Subcutaneous tissue	3	205.3			3	.058	312.6	652.3	529.8	184.2	111.6
Liver	2	165			3	.052	104.7	245	154.5	130.3	33.6
Kidney	2	193			3	.057	298	655	467	203	77

Pellets of testosterone propionate were implanted in the liver, as indicated in Table II. In this organ the pellet is not as effective as in the subcutaneous tissue. However, a moderate degree of androgenic activity is demonstrable. The pellet implanted in the kidney is as effective in stimulating the genital tract as the pellet in the sub-

cutaneous tissues. The average daily absorption from the pellet is approximately the same in all locations, as indicated in Table II.

Pellets of estrone[4] were implanted in the subcutaneous tissue, peri-renal fat, kidney, gastric omentum, liver or spleen of groups of castrated female rats, as indicated in Table III. In all animals except those having the pellet in the spleen, the smears were quickly converted to the estrus type and remained so for at least 60 days. For periods of 3 to 14 days after implantation of the pellet in the spleen, the smears of these animals contained leucocytes, epithelial cells and

TABLE III

Inactivation of estrone in castrated female rats

SITE OF IMPLANTATION OF PELLETS	NO. OF ANIMALS	TYPE OF SMEAR	
		Following implantation of pellet	Throughout remainder of experiment
Subcutaneous tissue	14	Estrous	Estrous
Spleen in normal position	8	Estrous 3–14 days	Anestrous
Spleen, later transplanted	4	Estrous 3–14 days; anestrous until transplantation	Estrous
Transplanted spleen	5	Estrous	Estrous
Gastric omentum	2	Estrous	Estrous
Liver	2	Estrous	Estrous
Kidney	1	Estrous	Estrous
Peri-renal fat	2	Estrous	Estrous

squamous cells. After this interval, the smear was composed of leucocytes, but occasionally some epithelial cells and a very few squamous cells were present among the leucocytes. In this group of animals daily observation showed these slight variations in the smear for periods of at least 70 days, and this almost constant leucocytic smear indicates that these animals were in an anestrous state.

In another group of castrated female rats, 21 days after implantation of the pellet of estrone in the spleen, this organ was transplanted to a site between the skin and abdominal muscles. The smear changed to an estrous type in from 3 to 14 days. Three weeks later, when the pedicle of the spleen was ligated, the smear was still of an

[4] Supplied under the synonym "Theelin" through the courtesy of Dr. Oliver Kamm of the Parke-Davis Company.

estrous character and remained so for more than 6 weeks. Pellets inserted into the previously transplanted spleen in a group of castrated female rats immediately produced estrus, which was maintained for more than 30 days. The pedicles were then ligated and the estrous smear remained unchanged for more than 50 days. In both these instances the pellets were situated in viable but altered splenic tissue. This was verified by microscopic examination.

Implantation of estrone pellets in the other sites (Table III) uniformly produces estrus. The amount of estrone absorbed daily, varies between 0.005 to 0.010 mg. in these locations, and the same rate of absorption is noted in the normally situated, and in the transplanted spleens.

One possible interpretation of these results is that testosterone propionate and estrone, implanted in the normally situated spleen, are inactivated as they are transported through the liver via the portal circulation. The less complete inactivation which takes place when the pellets are implanted directly in the liver substance may be explained by contending that part of the material is taken up by the hepatic venules before it is acted upon by the liver cells. However, the present experiments have not excluded the possibility that the spleen itself plays a rôle in the inactivation. When the spleen is transplanted and its pedicle ligated, certain changes occur in it which may interfere with its normal function.

Histological examinations were made of the spleens of the animals employed in these experiments. In those which were normally situated the structure of the pulp was normal. The pellets, whether testosterone propionate or estrone, were surrounded by a few fine strands of collagen with scattered fibroblasts and an occasional giant cell. The transplanted spleens with pedicle ligated were surrounded by a zone of dense fibrous tissue in which there were many small blood vessels. The pulp was altered by the presence of great numbers of large mononuclear cells, which also filled and choked the sinuses. The increased cellularity produced an apparent obliteration of the sinuses. The Malpighian corpuscles were normal in size and number, and often showed slightly increased activity in the germinal centers. In a few instances there were small zones of infarction; but the pellet was located in viable splenic tissue.

There is no way of judging whether these histological changes indicate an impairment of the inactivating power of the spleen. Therefore, the determination of the relative importance of the liver and spleen in the inactivating process in the living animal must await further study.

Conclusions: Pellets of testosterone propionate and estrone do not exert their specific effect on the appropriate castrate rats when implanted in the spleen. The specific effect returns when the spleens containing the pellets are transplanted and the splenic artery and vein are ligated. The rôle of the liver in this process is discussed.

The authors wish to express their appreciation to Dr. W. G. MacCallum for his kind interest and valuable criticisms.

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BOOK REVIEWS

(These reviews represent the individual opinions of the reviewers and not necessarily those of the members of the Editorial Board of this Journal.)

Textbook of Gynecology. By ARTHUR HALE CURTIS. 3rd ed. 603 pp. Illus. $7.00. *W. B. Saunders Company, Philadelphia, 1938.*

The author has added eight chapters to those in the preceding edition and has rewritten the entire book. These new chapters deal with gynecological anatomy and physiology, and with the endocrines as they affect the woman's reproductive system.

The anatomical descriptions are based on dissections of female cadavers carried out under the supervision of the Anatomical Department and they are of unusual interest. More space is given to anatomy than is usual in most gynecological textbooks.

The entire subject of gynecology is covered in a clear and interesting manner. The three-hundred and eighteen illustrations add to the value of the book.

Doctor Curtis' work is to be highly recommended as a textbook for students.

L. B.

Practical Dermatology and Syphilis. By HARRY M. ROBINSON. 397 pp. Illus. $4.50. *P. Blakiston's Son & Co., Inc., Philadelphia, 1939.*

This small book represents a very successful attempt to simplify the difficult subject of dermatology for medical students and practitioners. The arrangement of the book, which classifies skin diseases from the morphologic standpoint, includes a chapter on syphilis and then provides in brief though adequate form chapters on macular, papular, pustular, annular, ulcerative, excoriated, scarred and atrophic skin eruptions; and on those lesions involving the scalp, the bearded areas, the mouth, the sweat glands, and the nails. In each of these chapters are listed the several diseases which need to be considered in differentiation; and for each of these diseases there are brief statements concerning the sites of predilection, the objective and subjective symptoms, the etiology, therapy, and differential diagnosis.

The illustrations gathered from Dr. Robinson's many years of experience are excellent.

The book may be confidently recommended for medical students and physicians who will find it much more useful than many of the larger and more extensive texts. It should receive a wide distribution.

J. E. M.

Orthopedic Appliances. By HENRY H. JORDAN. 412 pp. Illus. $4.00. *Oxford University Press, New York, 1939* (Oxford Medical Publications).

Stressing the need for close cooperation between the orthopedic surgeon and the bracemaker, the author attempts to present the principles involved in the manufacture of supportive devices, in order to give "the orthopedist a better understanding of the limitations of bracemaking" and "the bracemaker a better understanding of the requirements of modern orthopedics." To accomplish this, he discusses the construction of a series of braces which, in general, were in use in the clinic of Hans von Bayer where the author had his orthopedic training. An enumeration of the materials used and of each step in the building of the more essential braces is given. Though there is no attempt to describe many of the braces in modern use, some of the devices described in the American journals are included. The book is well illustrated.

I. W. N.

Cranio-Cerebral Injuries. Their Diagnosis and Treatment. By DONALD MUNRO. 439 pp. Illus. $4.00. *Oxford University Press, New York, 1938* (Oxford Medical Publications).

This is a readable and instructive treatise. The increasing frequency of cranio-cerebral injuries is stressed and the author writes with a view to instructing the general surgeon to whom these patients come. The more complex procedures he wisely allots to the experienced neurosurgeon, located in the more fully equipped hospital.

Chapters dealing with fundamental cerebral physiopathology, with methods of history taking and examination, with injuries to the newborn, and with first aid are commendable in every respect. The numerous illustrative case histories are well presented. The frequent references to the underlying pathology are enlightening. The classification of injuries is essentially that which is in use elsewhere. The statistics of mortality and morbidity proclaim a high standard of therapy.

One gains the impression that the author has a trend toward the more radical types of treatment. He freely advocates the use of lumbar puncture for diagnosis, as well as treatment, of all types of injuries. In our experience this procedure is fraught with great danger and is far less effective in obtaining a permanent result than the safer subtemporal decompression; this is especially true when the injury is accompanied by an increase in intracranial pressure. The use of hypertonic intravenous injections, recommended by the author, has not proved effective in our hands. The use of x-ray films, as advocated, is of interest but such films are rarely of value except in the few cases in which subsequent operative procedures require precise localization of bony pathology. The author's method of treating compound fractures, including those which involve the paranasal sinuses, is more radical than would appear to be justifiable.

This book will serve to focus attention upon the methods of therapy employed

by the surgeons most concerned with the treatment of craniocerebral injuries and, for this reason, it will render a service by bringing to the fore a problem of ever increasing importance in modern life.

W. G. W.

Social Hygiene Nursing Techniques. By NADINE B. GEITZ. 82 pp. 25 cents and 5 cents postage. *American Social Hygiene Association, New York, 1938.*

This small booklet should be of value to clinic and public health nurses, social workers, and lay personnel engaged in venereal disease clinics. The information provided, which describes the procedures used in the New York City Health Department, is generally accurate and sufficiently detailed to fulfil its purpose.

J. E. M.

Diagnosis and Treatment of Diseases of the Thyroid. By JAMES H. MEANS AND EDWARD P. RICHARDSON. 367 pp. Illus. $5.00. *Oxford University Press New York, 1938.* (Reprinted from Oxford Monographs on Diagnosis and Treatment.)

The new features in this edition are not particularly significant. Most of the new knowledge about the thyroid gland during the past decade has had to do with this gland's interrelationship with other glands, and especially with the thyrotropic hormone of the anterior pituitary gland. Since, as yet, this knowledge does not affect diagnosis or treatment of diseases of the thyroid gland very little attention is paid to it in this book.

This new edition restates the work of the Thyroid Clinic of the Massachusetts General Hospital. Means has recently revised the section on Diseases of the Thyroid Gland for *Nelson's Loose-Leaf System* and in that article he considers the literature of the subject somewhat more broadly and comprehensively. However, it is useful to have the actual case histories to refer to and to realize that practically every statement in the book is based on the authors' personal opinion. This adds greatly to the readers' interest.

H. M. T., JR.

Immunity, Principles and Applications in Medicine and Public Health (Fifth Edition of "Resistance to Infectious Diseases"). By HANS ZINSSER, JOHN F. ENDERS, AND LEROY D. FOTHERGILL. 801 pp. $6.50. *The Macmillan Company, New York, 1939.*

Although this volume is published as the fifth edition of "Resistance to Infectious Diseases," it is actually a new book, completely rewritten and rearranged, and perhaps the most complete and most readable exposition of immunity yet to appear in this country. A deliberate attempt has been made to emphasize the applications of immunology to clinical and public health problems; and the authors have successfully incorporated the important contributions of the past ten years in their due relation to antecedent work. The very completeness and

excellence of that coverage make occasional lapses all the more obvious. Thus, if a reviewer may be permitted to ride a hobby horse, the essential physico-chemical mechanism and immunologic significance of the Wassermann and flocculation tests for syphilis are given scant consideration, while the complex charts showing the quantitative relationships of antigen, amboceptor, and complement, which are difficult to follow and are at best of dubious significance, are reproduced in almost a full-page illustration.

These, however, are but minor flaws in a volume which is a worthy successor to the previous editions. The student will find it an invaluable source-book; the teacher will find the arrangement and presentation pedagogically sound; and the investigator will be pleased with the clear and well-documented presentation of divergent views.

H. E.

A Synopsis of the Diagnosis of the Acute Surgical Diseases of the Abdomen. By JOHN A. HARDY. 345 pp. Illus. $4.50. *C. V. Mosby Company, St. Louis, 1938.*

Since the greater portion of abdominal surgery in this country is done by general practitioners, and since these men have not sufficient time to devote to concentrated study of the subject of abdominal diagnosis, the author has written this handbook of differential diagnosis of acute surgical diseases of the abdomen. No attempt is made to discuss operative procedures—the sole purpose of the book is to establish methods by which a diagnosis may be made without costly delay.

The first chapter deals with surface localization of the abdominal organs; the second stresses the importance of a general examination of the patient, rather than a concentration upon the abdomen alone. "An intuitive diagnosis is a rapid method of reaching a wrong conclusion." The third chapter stresses the importance of a complete and accurate history of the patient's illness with especial reference to pain, vomiting, and abnormalities in defecation and urination. A few brief, didactic statements are made about these symptoms but the physiological and pathological changes involved are not very thoroughly discussed. The remaining chapters take up in order acute surgical emergencies arising from diseases in the abdominal cavity.

The book is brief, well planned, and logical. It is particularly suitable when reference is quickly needed.

I. R. T.

The Vaginal Diaphragm. By LE MON CLARK, M.D. 107 pp. Illus. $2.00. *C. V. Mosby Company, St. Louis, 1939.*

This is a monograph devoted entirely to a discussion of one method of contraception: the vaginal diaphragm combined with a spermicidal jelly or cream. Dr. Clark quite properly emphasizes intelligent and skillful fitting and adequate instruction of the patient as essential to the successful use of this method.

The book is fully and excellently illustrated with photographs and sketches.

B. L. M.

Practical Microbiology and Public Health. By WILLIAM BARNARD SHARP. 492 pp. Illus. $4.50. *C. V. Mosby Company, St. Louis, 1938.*

This book is intended to accompany courses in "Preventive Medicine" or "Public Health," being given to medical students. The author presents the volume only as a handbook "designed to aid the student in organization, interpretation, and systematic record of data from laboratory and field." It is in no sense a textbook, nor does it do more than present the barest essentials, in synoptic form, of such a variety of subjects as general and clinical bacteriology and mycology; vehicles of infection; immunology; the methods used by the public health laboratory in examining milk, water, and foods, and in making various diagnostic tests; public health field surveys; garbage removal; ventilation; housing; restaurants; vital statistics; epidemiology; protozoology; helminthology; etc., etc. It is expected of the student that he be previously "familiarized by lecture and standard tests with the findings of authorities. . . ." The book is attractively printed and bound, and instructively illustrated. A number of laboratory exercises are detailed or suggested, and an index is appended.

K. F. M.

The Natural History of Population. By RAYMOND PEARL. 416 pp. $3.50. *Oxford University Press, London, 1939.*

In the University of London Heath Clark Lectures of 1937, Dr. Pearl presented some very fundamental contributions to the subject of human fertility and its relation to population problems. The publication of these lectures in book form under the title "Natural History of Population" makes available a body of extremely valuable material to the student of human biology. This material is varied in character, for it gathers together a large body of the published statistical evidence on human fertility, presents the results of Dr. Pearl's own investigations on the reproductive histories of nearly 31,000 women, and in addition contains a bibliography of about 700 titles pertaining to the subject of the book.

It should not be inferred from the large amount of statistical data presented that the book is merely a collection of tables. It is primarily a discussion and interpretation of the quantitative evidence on fertility. An analysis of the basic biological aspects of human fertility is followed by a discussion of its patterns as to such factors as age, race, and parity, and a more fully developed treatment of the extent and effects of contraceptive efforts. The final chapter deals with the implications of the various biological and environmental factors in fertility for the history and future course of world populations.

In this brief review it is not possible to summarize effectively the many interesting analyses presented in the text. They deserve the careful study of all investigators in the field of human biology, and will excite the interest of any person concerned with the behavior of mankind.

L. J. R.